Pathology

Handwritten Note

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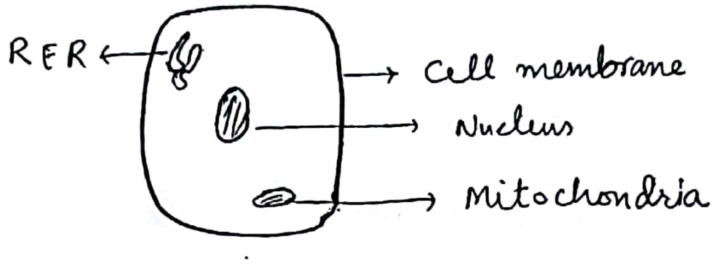
Name: ______________________

Subject: **Pathology**

Cell Injury

4 parts of a cell which are vulnerable to injury.



Causes of cell injury

1) Hypoxia (MC)*
   - (a) Ischemia* — Atherosclerosis / Thromboembolism / Vasospasm
   - (b) Anemia
   - (c) Cardiopulmonary failure.
   - (d) CO poisoning

2) Infections

3) Genetic disorders

4) Hypersensitivity reactions

5) Autoimmune diseases

6) Physical agents - heat, cold, trauma, radiation

7) Chemical agents

8) Nutritional imbalances — Deficiencies (Vit, PEM) / Excess (Vit ADEK, Fats)

Outcomes of cell injury

1) Irreversible cell injury / cell death

Necrosis   Necroptosis
Apoptosis   Pyroptosis.

2) Reversible cell injury :

Hydrophic degeneration (cloudy swelling)
Fatty denaturation

3) Cellular adaptation

Hypertrophy   Atrophy
Hyperplasia   Metaplasia

* Dysplasia is not a cellular adaptation. It is a premalignant condition.

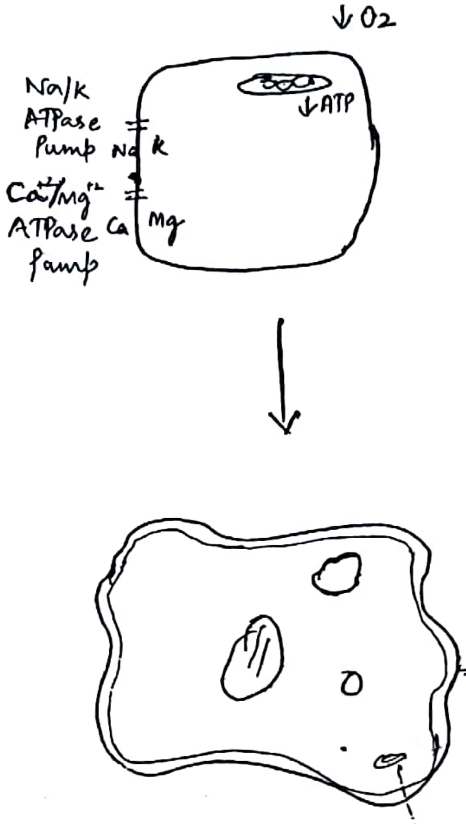
4) Intracellular accumulation & pathological calcification

Proteins
Fats
Glycogen
Pigments - Lipofuscin

Dystrophic cal.
Metastatic cal.

5) Cellular ageing

Mechanism of cell Injury (Reversible)



(a) Pump failure due to lack of ATP.

↳ Eflux of K from cell

↳ Na, Ca influx into the cell

↓

↑ Osmotic load

(H_2O enters the cell)

↓

Cellular swelling

Hydrophic changes

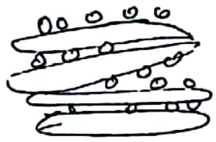
Organelles also swell up.

→ Cell membrane blebs are formed

→ Myelin figures are formed

↳ Due to accumulation of water b/w membrane phospholipids

(b) If ATP is absent - Protein synthesis stops.



↓ ATP

⟶

Dispersion of Ribosomes from RER

- Protein synthesis stops
- ↑ lipid synthesis & storage

⇓

Fatty changes

(c) Switch over to Anaerobic glycolysis

- Glycogen in cytoplasm is consumed
- Lactic acid is produced

⇓

↓ pH of the cell

Nucleus

↓ pH ⟶

Clumping of Nuclear chromatin

Mechanism of Irreversible cell injury

1) Severe mitochondrial damage

Important feature of irreversible cell injury:-

(I) ⇒ No ATP production by mitochondria

(II) ⇒ Large amorphous flocculant densities in mitochondrial matrix

↓

Composed of Calcium & phospholipids

2) Severe membrane damage

↑ permeability of plasma membrane

↓ leakage of important intracellular enzymes & proteins.

↓ Influx of Ca^{++} into the cell.

3) Release of lysosomal enzymes

↓ pH
↑↑ Ca^{++} in cytosol → Lysosome receptor irritation

↓

Acid hydrolase are released

- Phospholipases → Break membrane phospholipids
- Proteases → Break enzymatic & cytoskeletal proteins
- RNAses → Break RNA
- DNAses → Break the DNA in Nucleus

DNAses

↓

Cause random breaks in the DNA

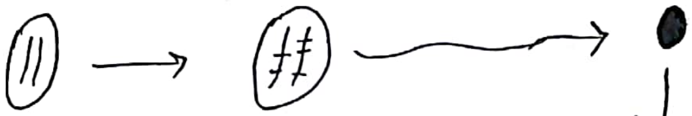
↓

Extract this DNA and do an agaros gel electrophoresis

↓

$\boxed{\text{Smear pattern}}$

Nuclear Changes



Pyknosis (condensation of chromatin)

↓

Karyohexis (fragmentation of nucleus)

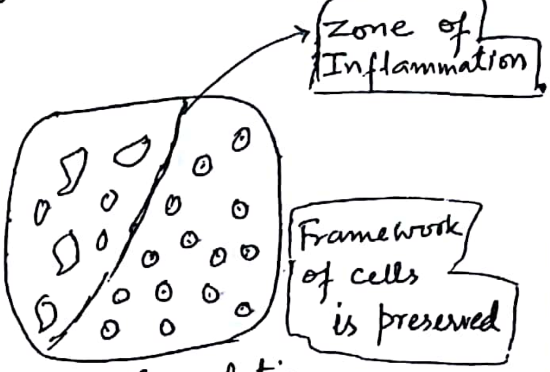
↓

Karyolysis (dissolution of nucleus)

H&E
- Stains Nucleus
- Stains cytoplasm & extracellular Connective tissue

Necrosis

Def. cell death in living tissue

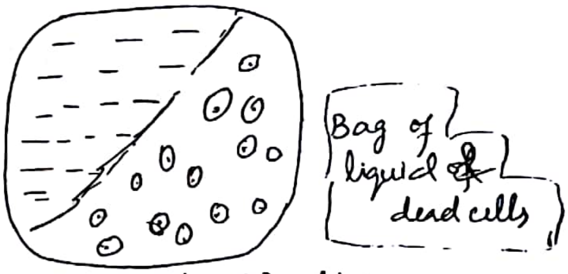


Coagulative

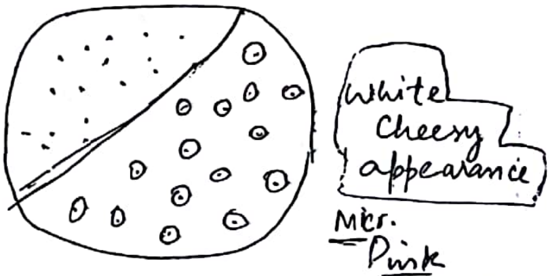
2 Processes that underlie necrosis are (Mech.)

ⓐ Enzymatic digestion of cells by lysosomal enzymes

ⓑ Denaturation of proteins



Liquifactive



Mcr. - Pink granular appearance

① Coagulative Necrosis

→ Due to denaturation of structural proteins of cells.

MC type of Necrosis

a) MC cause of Coagulative necrosis is Hypoxia except Brain

b) 2nd cause is Severe burns.

c) 3rd Dry gangrene.

d) Zenkers degenerations → Coagulative necrosis of muscle like rectus abdominis seen in pts with severe toxemia e.g typhoid

H/E staining

- Cell outline is preserved but cells do not have a nucleus
- ↑ cytoplasmic eosinophilia → due to denaturation of proteins
  → loss of blue nucleus
- Cytoplasm has a glassy appearance due to loss of glycogen
- Cytoplasm has a "Moth eaten" appearance due to loss of organelles.

M/c organ where coagulative necrosis is seen – HEART (MI)

Others – Liver
Kidney
Spleen.

Infarct

⇒ Area of necrosis, usually coagulative produced due to ischemia.

Infarcts are wedge or triangular shaped.

2 types

White Infarct (also called as pale/anemic Infarct)
Seen in solid organs with single blood supply
e.g. In Heart, kidney, spleen
Liver (in hypovolumic Shock)

Red Infarct (also called Hemorrhagic Infarct)
Seen in spongy organs
Organs with dual blood supply
Lung
Liver
(Red More common)

organs with collaterals
e.g Intestines
Torsion of Testis/ovaries.

2) Liquifactive Necrosis (colliquative Necrosis)

Due to enzymatic digestion of cells by lysosomal enzymes.

Dead tissue is converted into a bag of liquid.

e.g brain hypoxia (always Liquifactive Necrosis)

Abscess cavity — Bacterial / Fungal

Wet gangrene.

3) Caseous Necrosis

Dead tissue has a white, cheesy appearance and it is friable.

Miscroscopically – Pink granular appearance

Causes

M. Tuberculosis (Mycolic acid)

Syphilis

Fungal infections — Histoplasmosis / Coccidoidomycosis

4) Fat Necrosis

Enzymatic fat Necrosis

- Seen in pts with acute pancreatitis
- Abdominal cavity
  - ↳ Messentry
  - ↳ Omentum
  - ↳ Retroperitoneal fat

Lipase released from pancreatic acinar cells attack the fat cell membrane

FFA released from fat cell membrane

↓

FFA + Ca

↓

Chalky white deposits of fat necrosis

↓

Sponification of fats

Traumatic fat Necrosis

⇒ Breast

⇒ Subcutaneous tissue

↳ Due to trauma

5) Fibrinoid Necrosis

Areas of fibrinoid necrosis has a homogenous pink appearance due to deposition of fibrin [H & E Staining]

Causes

(a) Vasculitis e.g. PAN.

(b) Malignant HTN

(c) Peptic ulcers

(d) Aschoff nodules of RHD

Fibrinoid Necrosis in the centre surrounded by Caterpillar cells, L, PC

(e) AID → SLE → vasculitis
→ R A. → Rheumatoid nodules contain fibrinoid necrosis

Free Radicals

Molecules with unpaired electrons in the outermost orbit.

$O_2 \longrightarrow O_2^{\bar{\cdot}}$ (Superoxide)

$H_2 \longrightarrow H_2O_2 \rightarrow \boxed{OH^{\bar{\cdot}}}$ most Powerful free radicle

$Cl \longrightarrow HOCl^{\bar{\cdot}}$

$N \longrightarrow ONO\bar{O}$ (Peroxynitrite)

Causes of free radical generation

1. Normal Oxidation & reduction reaction occuring in the cells
2. Radiation ⟶ Hydrolysis → Free radicals
3. Oxygen toxicity.
4. Infections.
5. Drugs and chemicals.
6. Reperfusion injury → Heart, → Brain

Free radicals damage cells in 3 ways.

1. Lipid Peroxidation of membranes (Most imp. action)
2. DNA Damage (oxidative)
3. Oxidative damage to proteins in cytoplasm

Two reactions that generate ~~Peroxide~~ Free Radicals

Fenton's Rxn

$H_2O_2 + Fe^{2+} \longrightarrow OH^{\bar{\cdot}} + OH^- + Fe^{3+}$

Haber weiss Rxn

$O_2^{\bar{\cdot}} + H_2O_2 + H^- \longrightarrow O_2 + H_2O + OH^{\bar{\cdot}}$

Three enzymes that generate Free Radicals.

1. NADPH oxidases (Pagocytic oxidase)
2. Xanthine oxidase
3. Super oxide dismutase [Generates and also inactivates free Radicals]

Free Radical Removal Mechanisms

1. Antioxidants → Vit A, C, E
   Selinium
   Cystein & glutathione containing compounds.
2. Enzymes
   (a) Superoxide dismutase (SOD)-Inactivates Superoxides.
   - Cu Zn SOD (SOD I) [Found in Cytosol]
   - Mn SOD (SOD II) [Found in Mitochondria]

$O_2^{\bar{\cdot}} \xrightarrow{SOD} H_2O_2 + H_2O$

Generates H_2O_2.

(b) Catalase

Found in peroxisomes

Inactivates H_2O_2

Peroxisomes are organelles where free radicals are generated and inactivated.

(c) Glutathione peroxidase → Cytoplasm → Mitochondria

Inactivates — H_2O_2 — $OH^{\cdot}$

③ Serum Proteins

Albumin ⟶ binds Fe & Cu

Lactoferin ⟶ Fe binding

Transferrin ⟶ Fe binding

Ceruloplasmin → Cu binding.

⇒ Free radicals can cause cell death by

- Necrosis
- Apoptosis
- Necroptosis

Brain is protected from Free Radical injury by CuZn SOD (SOD I)

$\xrightarrow{\text{Mutation}}$ Amyotropic Lateral Sclerosis of Brain

Most important Ion involved in cell injury - Ca^{++}

First ion involved - Na

Most susceptible to ischemic injury - Neurons (3-4 min) followed by Cardiac tissues (20-40 min)

Tissue least susceptible to ischemic Injury - Fibroblasts followed by Skeletal muscle

First sign in all types of cell injury except apoptosis - Hydrophic Change

Apoptosis

- (Programmed cell death)
- Active process.
- Death of single cell or small group of cells.
- No surrounding inflammation around apoptotic cells.

e.g; Physiological

→ Embryogenesis

→ Involution of hormone dependent organs upon hormonal withdrawl

⇒ Breakdown of endometrium during mensturation

⇒ Ovarial follicular atresia following menopause

→ Cell death in rapidly proliferating cells like cells in GIT, Skin, Respitatory tract.

→ Death of cells that have served their purpose e.g death of neutrophills at the end of acute inflammation.

→ Elimination of harmfull self reactive lymphocytes

Pathological

- DNA damage
- Accumulation of misfolded proteins
- Viral infections
  - Adenovirus/HIV
  - Viral hepatitis

    ↓

    Councilman bodies
    (Apoptotic hepatocytes)
- Duct obstruction of organs produces atrophy and apoptosis
  e.g: Salivary gland
  Pancreas
  Kidney (ureter obstruction)

Morphology

2 enzymes that bring about apoptosis are

Caspases

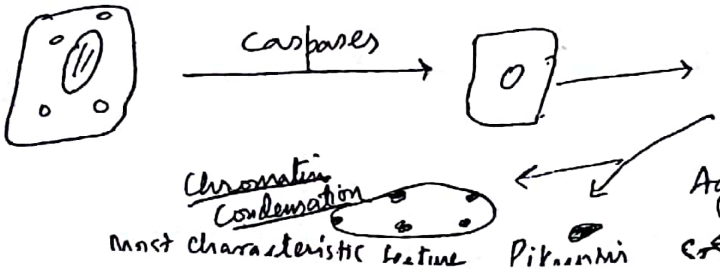
Endonucleases (Ca, Mg dependent enzymes)

Caspases break the framework of the cell → cell will shrink & tight packing of organelles

Endonucleases break DNA at specific sites (Internucleo-somal Regions)

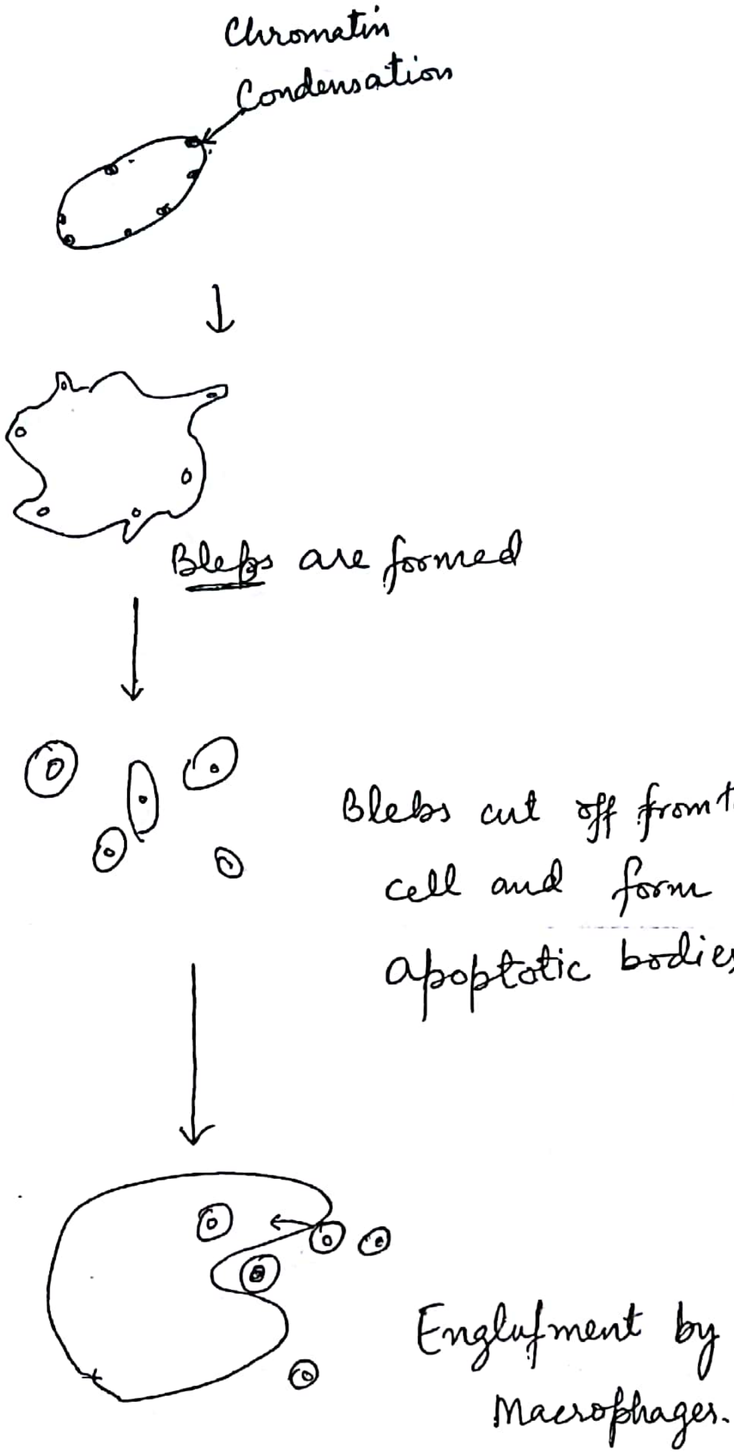
↓

180 - 200 bp DNA fragments are produced



Agarose gel ← electrophoreses

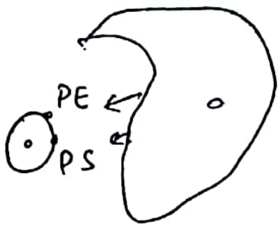
STEP LADDER PATTERN



Apoptotic bodies are removed by Macrophages because.

⇒ Apoptotic bodies express **Thrombospondin I** on their outer leaflet and macrophages have receptor for Thrombospondin II (absent in normal cells)

- Apoptotic bodies express phosphatidyl serine (PS) and phoshatidyl ethanolamine (PE) on their outer surface & Macrophages have receptors for PS & PE



Annexin V is a marker of cells undergoing apoptosis.

It is an immunostain that stains PS & PE which are expressed on outer leaflet of apoptotic body.

Normal cells have PS & PE on Inner leaflet thus remain unstained by Annexin IV

Microscopically
- Shrunken
- Hypereosinophilic
- Chromatin condensation below the nuclear membrane / Pyknosis in nucleus.

Pathways of Apoptosis

1) Intrinsic/ Mitochondrial Pathway
2) Extrinsic/ Death receptor initiated Pathway
3) p53 Pathway
4) Perforin granzyme dependent killing.

① Intrinsic /Mitochondrial Pathway

Brought about by BCL 2 family of gene.

20 members in the family.

- Antiapoptotic members (Prevent apoptosis)
- Proapoptotic members (Lead to apoptosis)
- Arbiters (Regulate balance)

Antiapoptotic members

BCL2 BCL-XL, MCL-1

They reside in the outer mitochondrial membrane ER membrane & cytosol

↓

Keep the permeability of the membranes Intact

↓

Prevent leakage of Cytochrom C ⇒ No Apoptosis

Proapoptotic members.

BAX BAK

They increase the permeability of outer mitochondrial membrane (drills holes in OMM)

↓

Leakage of cytochrome C from mitochondria into cytosole

↓

which activates $\boxed{\text{Apaf-1}}$ (Apoptosis activating factor-1)

↓

Apoptosome is formed.

↓

Binds and activates Pro Caspase 9.

⇓

Apoptosis

Arbiters of apoptosis

BIM BID BAD PUMA NOXA

Also called as [Sensors of cellular stress] & [BH3 only proteins]

Function is to regulate balance b/w above two groups

Inhibitors of intrinsic pathway

(IAP → Inhibitor of apoptotic pathway proteins)

↓

Inhibits Procaspase 9

Smac/Diablo – Mitochondrial proteins.

Pro apoptotic

They Inactivate IAP

↓

Thus promoting Apoptosis

Caspases

Initiator Caspases

Caspases 8, 9, 10

Intrinsic → Caspase 9

Extrinsic → Caspase 8, 10

Executioner Caspases

Caspases 3, 6, 7

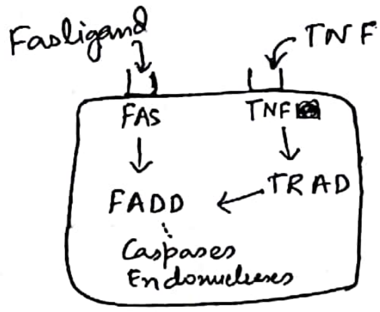
Most important

Common for both the pathways

Extrinsic/Death receptor initiated pathway.

Death receptors start this pathway
↳ Belong to TNF receptor family

FAS (CD95) ← TNF receptor family → TNFR1



FADD → FAS associated Death domain

TRAD - TNF Receptor 1 Associated Death domain.

Inhibitor of extrinsic pathway

FLIP → Inhibits procaspase 8

Produced by viruses & normal cells.

p53 Pathway

Tumor suppressor gene

Chromosome 17p 13.1

Called as :- (Molecular Policeman)
(Guardian of Genome)
(Critical gate keeper.)

It applies Emergency breaks and causes G1 arrest of cells.
~~genome on~~

Cells with damaged DNA enter the cell cycle

↓

Sensors of DNA damage ~~of~~ are activated.
(ATM and RAD family of proteins)

↓

Sensors activate transducers
(CHEK kinase family of proteins are transducers)

↓

p53 activated

↓

p53 recruits p21 (Inhibitor of cyclins & CDKs)

↓

p21 causes G1 arrest of cells.

⇓

Now p53 assesses DNA Damage

Too much DNA damage

↓

Induces Apoptosis via mitochondrial pathway by <u>BAX</u>.

Little DNA damage

↓

DNA repair gene GADD45 is recruited & DNA repair done.

$t_{1/2}$ of p53 → 20 minutes

MDMX

MDM2

Loss of p53 (both copies) → Li Fraumen's syndrome

↓

↑ risk of developing carcinomas, Sarcomas, lymphomas, etc.

Gene most commonly mutated in humans carcinoma is p53.

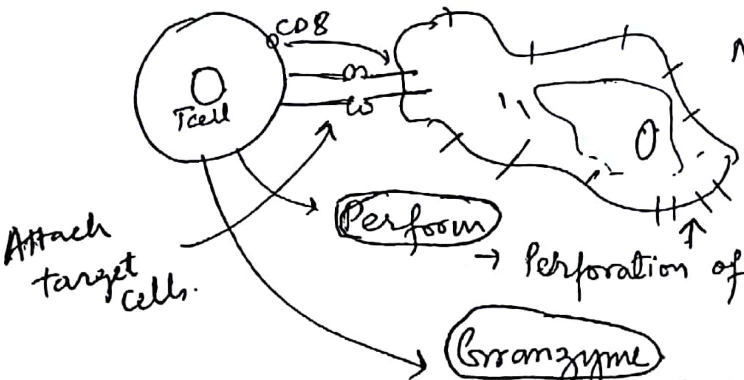
Perforin Granzyme Dependent killing

Pathway used by NK cells and Cytotoxic Tcells (CD8 cells)

⇓

Induce apoptosis of target cells.

↓ Virally infected cells & Tumor cells.



Perforin → Perforation of cell membrane

Granzyme → activates Caspases & endonucleases & leads to apoptosis

Autophagy - cell eats its own contents
Seen in ~~nutrient~~ nutrient deprivation.

Membrane of autophagic vacoule is derived from ER

Autophagic vacoule fuses with lysosome
Lysosomal enzymes degrade the contents → used as source of nutrient.

LC 3 is a marker of cells undergoing autophagy
(Light chain) Microtuble associated
→ Identifies targets of autophagy
→ Formation of autophagic vacuole.

Differences b/w Necrosis & Apoptosis

Necrosis	Apoptosis
- Passive process	Active process.
- Death of large no. of cells or large parts of the organ.	- Death of single cell or small groups of cells.
- Cells swell up	- Cells shrink.
- Cell membrane permeability is increased	- Cell membrane permeability remains intact.
- Surrounding Inflammation is present	- No surrounding Inflammation is seen.
- DNA is broken by DNASES ↓ Smear pattern	- DNA is broken by endonucleases ↓ Step ladder pattern
↳ Pyknosis ↳ Karyorhexis ↳ karyolysis	Pyknosis < Chromatin condensation below the nuclear membrane.
- Pathological process	- Both physiological & pathological.

Necroptosis

Programmed cell death

Active process.

Also called ~~protoso~~ Programmed Necrosis

Hybrid of Necrosis and apoptosis.

↳ Starts similar to extrinsic pathway

⇓

TNF + TNFR 1

↓

Multiprotein Complex called **Necrosome** is formed.

[Necrosome → RIP 1, RIP 3 & Procaspase 8]

RIP

[Receptor Interacting Protein kinase]

~~Procaspase~~

↓

Necrosome (Procaspase 8 is not activated)

↓

Metabolic alteration in cell

↓

↓ ATP ← → ↑ ROS

↑ ROS → DNA & protein damage

↑ ROS → Membrane damage (by lipid Peroxidase)

↓ ATP, DNA & protein damage, Membrane damage → ↑ membrane Permeability & leakage of contents

⇓

Cell death like Necrosis

Examples of Necroptosis.

Physiological – Occurs during formation of bone growth plate.

Pathological – Cell death in Steatohepatitis, acute pancreatitis, reperfusion Injury, & Neurodegenerative diseases e.g. Parkinson's disease.

Pyroptosis

Active process
Programmed cell death.
Also called as pyrogen induced apoptosis.
[accompanied by fever]

→ Receptors of innate immunity
→ Microbial products are added
→ Inflamasome (multiprotein product)
↓
Activates Caspase I AKA (IL 1β converting enzyme)
↓
IL 1
↙ Fever
↘ Inflammation.

⇒ Caspase I & Caspase II induce cell death similar to necrosis

Cellular Adaptations

① Hypertrophy

↑ size of cells → ↑ size of organ
↓
↑ in functional capacity of the organ.

Size of cell increases due to increase in proteins and organelles in the cell.

Cause → ↑ Functional demand
→ ↑ Growth factor / Hormonal stimulation.

Physiological

⇒ Gravid uterus increases in size due to estrogens.
Hypertrophy >> Hyperplasia.

⇒ Skeletal muscles in weight lifters.
⇒ Lactating breast ↑ in size due to Hypertrophy.

Pathological

⇒ Cardiac hypertrophy / LVH due to HTN or aortic valve disease.

Different isoform of myosin in hypertrophic heart

Switch of contractile protein from adult (α myosin) to fetal form (β myosin) which produces slower and energitically economical contractions.

$\alpha \rightarrow \beta$ myosin

(2) Hyperplasia

↑ in no. of cells ⟶ ↑ in size of organ
↓
↑ in functional capacity.

Physiological

Hormonal Hyperplasia
Seen in breast during puberty & pregnancy

Compensatory Hyperplasia
seen in liver.

Pathological

Due to excessive hormone or GF stimulation.

Endometrial Hyperplasia
due to excessive estrogen

BHP
due to androgen.

Pathological hyperplasia can lead to carcinoma.

Endometrial hyperplasia ⟶ Carcinoma

⇒ Q Endometrial adenoca can arise in background of endometrial hyperplasia

⇒ Q Endometrial adenoca can arise in background of endometrial atrophy also.

(3) Atrophy

↓ in size of cells (due to loss of proteins & organelles)

↓

↓ size of organs

↓

↓ in functional capacity of organ.

Causes
- ↓ work load
- ↓ Loss of Nerve supply
- ↓ blood supply
- Inadequate Nutrition
- Loss of hormonal stimulations
- Ageing (Senile atrophy)

Atrophic Cells → ↓ Protein Synthesis

Atrophic Cells → ↑ protein degradation by ubiquitin proteasome pathway

Atrophic Cells → Show autophagy.

Example Brown atrophy of heart.

[Atrophy + Lipofuscin accumulation]

(4) Metaplasia. One type of adult cell is replaced by another type of adult cell because another one is better suited for the environmental condition.

- Due to stem cell reprogramming.

e.g.: (a) Columnar to squamous metaplasia seen in smokers & Vitamin A deficient patients (Respiratory Tract)

Protective function of epithelium is lost.

(b) Squamous to columnar metaplasia in Barrett's esophagus in pt of GERD.

(c) Mesenchymal metaplasia → Bone is formed in soft tissue in foci of injury e.g.: Myositis ossificans.

Stain used is Mucin

Intracellular accumulations and Pathological Calcification

Pathological Calcification

Dystrophic Calcification

- Seen in dead/damaged tissue.
- Serum Ca. N.
- Ca metabolism N

e.g – dead parasites
- areas of Necrosis
- damaged heart values (as in RHD)
- Blood vessels – Atherosclerotic plaques
  – Moncke berg's medial Calcificat sclerosis
  Calcification in media of medium sized arteries in old ages
- Psammoma Bodies – Concentric calcification on necrotic tumor cells.

Metastatic Calcifications

- Seen in normal tissues due to hyper-calcemia
- Serum Ca ↑
- Ca metabolism deranged

Sites

MC ① – Alveolar Septae of lungs
② kidney
③ Gastric mucosa
④ Walls of systemic arteries and pulmonary veins

Causes

① CRF
② Hyperparathyroidism
③ Vit D intoxication
④ Milk alkali syndrome.
⑤ Sarcoidosis
⑥ Pagets diseases
Bone diseases
⑦ Bone tumor (M.M).

* Calcification starts in mitochondria of all cells except kidney, where it starts in the basement membrane of tubules.

Stains for Calcium → Von Kossa (Black)
→ Alzarine Red S (Red)

Calcein is another stain for calcium.

Tetracycline labelling index is done to detect bone mineralization.

Intracellular accumulations.

① Proteins
→ Russel bodies in Plasma cells in M.M.

→ Intracytoplasmic accumulation of Ig in PC cytoplasm

→ Resorption droplets in PCT cells in nephrotic syndrome

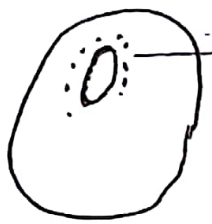
② Fat
→ Triglyceride accumulation → Fatty liver.
→ Cholesterol accumulation → Xantomas
→ Atherosclerotic plaques

③

③ Glycogen
→ glycogen storage diseases
→ seen in PCT cells of kidney in Pts c̄ DM → ARMANI EBSTEIN Lesions.

④ Pigments

→ Lipofucin
Lipochrome pigment
Brown in colour
↳ Pigment of ageing
↳ Wear and Tear pigment.
↳ Sign of Free radical injury to cells.



→ accumulates in perinuclear in the lysosomes in the cytoplasm.

→ Hemosiderin
Iron containing pigment
→ Stain - Perl's prussian Blue reaction
H & E - golden brown in colour
Accumulation in conditions of Iron overload.
e.g severe hemolytic anemia (Thal major)
Areas of hematoma
Hemochromatosis

Cellular Ageing

Telomeres Ends of chromosomes by which chromosomes are attached to nuclear membrane.

Also called as Biological clocks because with each cell division there is telomere shortening And when telomeres are shortened beyond a critical limit → Cellular Senescence
(terminal non dividing state of cell)

Telomerase Maintains the length of telomeres.

Not found in somatic cells

Found in
- Stem cells
- Germ cells
- Embryonic cells
- Cancer cells.

Telomere lengthening → Carcinogenesis

90% human cancers are +ve for telomerase enzyme

Werner's Syndrome → Premature ageing
Def. of enzyme DNA helicase
↓
Needed for DNA unwinding & repair.

Calorie restriction promotes longivity!

Calorie restriction promotes longevity

↑ Sirtulins (SIR)

↓ Signaling through IGF pathway
↓
↓ cell growth &
↓ cell metabolism
↓
↓ cell damage.

<u>Sirtulins</u> NAD dependent protein deacytylases.

Distributed in different components of cell

7 different types known (SIR 7)

⇒ Prevent free radical damage
↓ Apoptosis
Stimulate protein folding
Inhibit cell metabolism

} Promotes Longivity

Associated with
- Ageing
- DM
- Cancer.

Sirtulins & Lipofucin

Tissue processing

[Paraffin Embedding] (10-12 hrs)

Steps

① Fixation

MC fixator used is 10 % Neutral buffer formalin

② Dehydration

Putting tissue in ↑ grades of ethyl alcohol.

(30% ethanol → 50% → 70% → absolute alcohol)

③ Clearing

- Makes the tissue optically clear.
- Clearing agent is miscible with paraffin wax.

MC Xylene

④ Embedding

Molten paraffin wax enters the tissue and it hardens the tissue

⑤ Sectioning

Cut 4-6 microns thin sections and place them on slides → Stain $\bar{c}$ H&E.

Fixatives

① M/c used fixative for HPE – 10% neutral buffered formaline.

② EM → Fixative is 2.5% glutaraldehyde
↓
Followed by post fixation in Osmium Tetraoxide
↓
Ultra thin sections are cut (1-2 micron)

Stain → Uranyl acetate
& observed in EM.

③ PAP smear fixed in 95% ethanol (Absolute alcohol)

④ IF examination → fixed in Normal Saline
↓
Frozen section is cut
↓
Section placed on slide
& stained by IF stains &
↓
Observed in UV light.

(5) Fixatives for Peripheral Smear and FNAC.
↳ Methanol.

Uses of Frozen Section

Also called as Cryosection
or
Intraoperative biopsy

Tissue is frozen at -20° to -30°C in a cryostat.

Frozen section (1) ⇒ Can descriminate benign from malignant

(2) ⇒ Can analyse resection margins.

(3) ⇒ Can detect metastatic deposits in sentinal nodes.

(4) ⇒ Used for demonstration of fat in tissue.

(5) ⇒ Immuno fluoresence examination
- Tissue cut by frozen section.

Special Stains

① MC stain used is H & E

② Fat — Oil Red O (Red)
— Sudan Black
— Orcein

③ Glycogen — PAS

Colour - Pink/Rose Pink

→ glycogen is PAS +ve and diastase sensitive.

PAS +ve substances

Amyloid ⇒ PAS +ve diastase resistant.

- Glycogen
- Glycolipids
- Glycoproteins
- Mucins
- Colloid
- Amyloid
- Basement membrane
- All fungi
- Russel bodies.

GGG CAMBRF

Used in Diagnosis of

- Glycogen storage disorder
- Staining of macrophages in Whipple's disease
- Demonstrating fungi
- Mucins in adenocarcinoma of large intestines.
- Seminoma, rhabdomyosarcoma, Ewig's Sarcoma — contain glycogen
- Lymphoblasts in AML (Block PAS +ve)

(4) Mucins (glcosylated proteins - plasma membrane, mucosas membrane)

3 Stains
- Alcian blue
- PAS
- Mucicarmine - Magenta
  - Can stain crytococcus Capsule also only not the whole fungi, (Red coloured)

(5) Basement membrane Stains

- PAS
- Silver Stain (Black colour)

(6) Connective tissue Stains

- collagen - Massons Trichrome → Blue colour
- Reticulin fibre - Silver Stain ↳ Black
- Elastic fibers - Verhoff stain

(7) Fibrin & Muscle

PTAH (Phospho Tungstic Acid Hematoxylin)

(8) Fat/lipids

Oil Red O, Sudan black (Best) & Orcein

(9) Calcium

Von kossa (Black)
Alzarin Red S (Red)
Calcein

(10) Melanin pigment

Masson Fontana — BLACK (Silver Stain)

Dopa reaction.

(11) Stain for Copper

Rubeanic acid

Rhodamine

Orcein

(12) Hemosiderin pigment

Perl's Prussian Blue reaction

↓

Hemochrometosis

(13) Bile pigment

Fouchet's Technique (green)

(14) Mast cells

Toluidine Blue

Frozen section - Toluidine Blue.

(15) Stains for Microorganism

(a) Mycobacteria Zeihl Neelsen Stain

(b) Lepra Bacilli Fite stain.

(c) Fungus PAS (Dead only)

GMS (Gomori Methamine Silver) → Best; → Live & Dead

(d) Spirochetes Warthin Starry (Silver stain) for H pylori

Inflammation

- It is Response of vascularized connective tissue to injury.
- Protective Injury

Acute	Chronic
Exudate is formed	

Exudate	Transudate
- Inflammatory edema	Non Inflammatory edema
- Formed due to increased vascular permeability	- Formed due to increased hydrostatic pressure.
- Rich in proteins & cells	- Poor in proteins & cells.
- Sp. gravity >1.020	- Sp. gravity <1.012
- LDH ↑	- LDH ↓

Acute	Chronic
(1) Sudden onset	(1). Insideous onset.
(2) Lasts for a short duration	(2) Lasts for long duration (weeks- months- years)
(3) 2 characterstic features - extravassation of neutrophils - exudate formation	(3) 2 characteristic features - infiltration of tissue by mononuclear cells - Tissue destruction.
(4) Local signs & symptoms are prominent.	(4) Local signs & symptoms are not prominent.
(5) Usually self limiting	(5) Progressive
(6) Tissue injury is mild.	(6) Severe & may lead to fibrosis.

Acute Inflammation

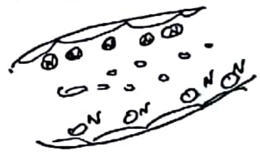
- Events of acute inflammation
- Mediators of acute inflammation

Events of acute inflammation.

- Seen in post capillary venules
  - → Vascular events
  - → Cellular events.

(I) Vascular events

(1) Transient vasoconstriction
↓
(2) Massive vasodilation
↓
(3) Increased vasular permeability. → exudate formation (Most imp)
↓
(4) Stasis of cells in Blood vessels.



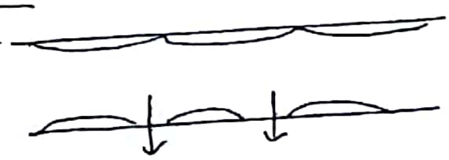
↓
(5) Leucocyte margination to the periphery.

(II) Cellular events

- Adhesion & transmigration
- Chemotaxis
- Phagocytosis

Mechanisms of ↑ vascular permeability

(1) Endothelial contraction (M/c mech)
(occurs in post capillary venules)



Mediators →
- Histamine
- Bradykinin
- Substance P
- Leukotriene

⇒ Immediate Transient response

② Endothelial retraction / Junctional reorganisation

Delayed type of contraction (vennules & capillaries)

Mediators (cytokines) → IL-1
→ TNF α

⇒② Delayed, sustained response

③ Direct Injury

(Seen in venules, arterioles, capillaries)

Severe Injury

e.g Severe burns
Chemicals
Toxins

↓

Cells undergo necrosis & detach.

↓

Fluid leakage which starts immediately

↓

Leakage continues till a new cell regenerates

⇒ Immediate sustained response

Mild Injury

e.g mild sunburn.

↓

endothelial cells die after a few hours due to apoptosis.

↓

Fluid leakage starts after few hours

↓

Leakage continues till a new cell regenerates

⇒① Delayed sustained response

Transplant Rejection

For a transplant HLA matching is done b/w donor and receipient.

HLA antigens that should be matched are.

HLA DR

HLA B

HLA A

HLA matching is not done for liver, heart, lungs, & cornea

because other factors like

- anatomical combatibility
- Severity of underlying disease
- Need to minimize the ~~time~~ time of organ storage

Immunosuppressive therapy

1. Steroid - Reduce Inflammation
2. Mycophenolate mofetil - Inhibits lymphocyte proliferation
3. Tacrolimus (FK 506) - Inhibits Phosphatase Calcineurin which is required for activation of NFAT (Nuclear Factor of Activated T cells)

NFAT not activated
↓
No IL2
↓
T cell inhibition

(4) T & B cell depleting antibodies

(5) Pooled IV immunoglobulins.

Complication of organ transplant.

(1) Infection (MC)

→ CMV most common infection
Cells show owl eye intranuclear inclusions.

→ Polyoma BK viral infection
Decoy cells - PCT cells $\bar{c}$ intra-nuclear basophilic inclusion.

(2) Transplant rejection

(3) GVHD

(4) ↑ risk of malignancies
→ HPV associated SCC skin
→ EBV ass. lymphomas
→ HHV8 ass. Kaposi's Sarcoma.

Transplant Rejection

Solid organ transplant

Recipient is immunocompetent.

3 types of Rejection.

① Hyperacute rejection
occurs within minutes
(< 48 hours)

It is due to preformed antibodies in the recipient.

↳ seen in
- Multiparous women
- Multitransfused individuals
- Past H/o transplant rejection.

* ABO & RH incompatibility can also cause hyperacute rejection.

Gross

Slightly swollen + mottled + cyanosed

kidney — Anuric
Filters few drops of blood urine or no urine at all.

M/E

① Neutrophilic infiltration in glomerular capilaries, arterioles & peritubular capillaries

② Fibrinoid necrosis & thrombosis in the vessels

③ Thrombosis leads to cortical necrosis

TYPE II Hypersensitivity Reaction.

Acute Rejection

Occurs from weeks - months (< 6 months)
or
later when immunosuppressive therapy is withdrawn.

2 types

Acute cellular Rejection

Brought about by
- CD4 T cell
- CD8 T cell

Type IV HR.

Responds to increasing the dose of immunosupressive therapy.

Acute Humoral Rejection

Brought about by antibodies that are produced after transplantation.

Type II > Type III HR.

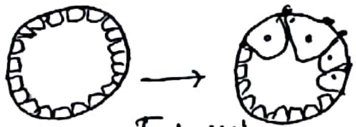
Doesnot respond to increasing the dose of immuno-supressive therapy but responds to B cell depleting agents.

Biopsy
↓
Microscopical Ex

Three types are seen.

Type I / Tubulointerstitial pattern
- Tubulitis
- (T cell/macrophage) Interstitial mononuclear cells Infiltration

M/C
Vasculitis
↓
Damage to glomeruli & small vessels



Tubulitis

Type II vascular pattern
Endothelitis

Type III vascular pattern
Endothelitis
+
Necrosis in the vessel wall.

Thrombi in small vessels

Fibrinoid Necrosis in vessels

IHC - C4d deposited in vessels
- glomerular Capillaries
- Peritubular Capillaries.

[in immunoperoxidase Stain]

Chronic Rejection

(Months - Years)

Can be due to antibodies or Tcells

More common - Type IV HR.

M/E - ① Obliterative intimal fibrosis of BV.
② Atherosclerosis of graft vessels
③ Glomerulosclerosis
④ Tubular atrophy
⑤ Intersititial fibrosis

Transplant glomerulopathy.

GVHD

Graft Versus host disease

It is a complication of Bone Marrow Transplant. (BMT)

Type IV HR.

HSC
TL

BMT ← Hematopoietic S. cells (HSC)
Donor T lymphocytes

Recipient is immunocompromised

- Due to primary disease for which Transplant is indicated
- Due to Radiotherapy or chemotherapy that is given to form transplant bed.

GVHD

Acute (<100 days)

1. Skin (MC)
   Rashes & ulcers
2. GIT Oesophageal & Intestinal ulcers
   Bloody diarrhea
   Malabsorption

Chronic (>100 days)

1. Skin
   Sclerodema like fibrosis
2. Strictures
   Malabsorption.

③ Liver

Cholestatic jaundice — Destruction of the ducts
Inflammation of bile duct — Cholestatic Jaundice

④ Destruction of immune system of the recipient
e.g: LN, Thymus, spleen, etc.

T cell depleted B.M.T.

- No GVHD
- Increased incidence of graft failure
- Relapse of primary disease eg. Acute leukemia.

Also called as graft versus leukemia effect.

- ↑ incidence of EBV. related leukemias & lymphomas.

Neoplasia

- New growth
- Neoplasm - Tumor

Benign

Suffix "oma"

Epithelial origin → Adenoma
→ Papilloma

Mesenchymal origin ⇒
Lipoma
Fibroma
Leiomyoma
Rhabdomyoma
Osteoma
Chondroma

Malignant

Suffix carcinoma/sarcoma

Epithelial origin → Carcinoma
e.g. Adenocarcinoma
SCC
Transitional Ca
BCC

Mesenchymal origin → Sarcoma
e.g. Liposarcoma
Fibrosarcoma
Leiomyosarcoma
Rhabdomyosarcoma
Osteosarcoma
Chondrosarcoma.

Malignant tumors c̄ Suffix 'Oma'.

1. Hepatoma (HCC)
2. Seminoma
3. Melanoma (melanocytes)
4. Chordoma (Malignant tumor of Notochord)

Teratoma

Tumor of Totipotent cells

(a) Mature cystic teratoma [Benign tumor]

→ Elements of all three germ layers

→ All components are mature.

e.g.: Skin adnexal structures, bone, cartilage, blood vessels, tooth, fat, thyroid tissue

ovary → Dermoid cyst.

(b) Immature Teratoma [Malignant tumor]

Elements are immature or fetal type

e.g. immature cartilage or bone

immature Neural tissue is important

↳ for grading.

(c) Teratoma with malignant transformation

Malignat tumor

One of the component has become malignant.

Pleomorphic adenoma

- Benign tumor
- Salivary glands (M/c)
  (Parotid MC
  & Breast)

Epithelial component
- glands
- Sq. epithelium

Mesenchymal component
- Cartilage
- Fibrous tissue
- Bone
- Muscle

Cells of origin → Myo epithelial cell.

Rx Wide excision

As Tongues of tumor tissue are seen in the surrounding tissue.

NOT TRUE NEOPLASMS

Choristoma

- Ectopic rest of Normal tissue
- Normal tissue at abnormal location

e.g glial tissue in Nasopharynx
Adrenal rests in the kidney

Hamartoma

Normal tissue in normal location but in a disorganised arrangement.

e.g bronchial hamartoma.
Hemangioma
Lymphangioma.

Benign	Malignant
Well differentiated (Resemble the tissue from which they rise)	Poorly differentiated or well - moderate differentiated or Anaplastic ↓ (Complete lack of differentiation)
Slow growing & may regress on their own	Rapidly growing, have erratic growth.
Usually Encapsulated. Un encapsulated are as - Leiomyoma, Hemangioma, Lymphangioma	Malignant tumors that show spontaneous regression - RCC - Neuroblastoma - Retinoblastoma - Malignat Melanoma - Chorio Carcinoma.
	Uncapsulated or may show pseudo capsule (formed by compressed normal tissue)
No local invasion	Local invasion +
No metastasis	Metastasis +

Metastasis is most imp. feature that differentiate ~~that~~ bening from malignant

Malignant tumors that show local invasion but no metastasis ⟹ BCC
Glioma

Features of Malignant cells

Large cells
High N:C ratio
Pleomorphism - variation in shape & size
Hyperchromatic nucleus
Abnormal mitosis

Dysplasia

Premalignant condition e.g. Cervical dysplasia

Loss of polarity
Pleomorphism
High N:C Ratio
Abnormal mitosis
Hyperchromatic Nucleus

They are confined within the basement membrane

Cervical dysplasia

CIN I - Abnormal cells confined to 1/3rd of Cx

CIN II - occupy 2/3rd Cx epithelium

CIN III - occupy full thickness of cervical epithelium | BM intact.

Pathways for Spread.

① Lymphatic Spread

Preferred mode of Spread for a Carcinoma

LN drainage Ca. → Metastasis

LN drainage Ca. → Reactive hyperplasia

② Hematogenous Spread

Preffered mode of spread of a Sarcoma

3 carcinomas first spread by Hemato-genous route:
- RCC
- HCC
- Choriocarcinoma

M/C Spread to - lungs > Liver

Venous invasion is more common than Arterial invasion

③ Direct Spread

(a) Mesothelioma (MC) arises from mesothelial cells of pleura or peritoneium & spreads on the surface of pleura or peritoneum.

(b) Pseudomyxoma peritonei

Seen in
- Mucinous adenoCa Appendix
- Mucinous adenoCa ovary
- Mucinous adenoCa Colon

Mucin produced by tumor cells
↓
Causes adhesions b/w various organs in the peritoneal cavity.

Tumors that spread via CSF

Medulloblastoma

$\boxed{\text{Small round cell tumor}}$

$\boxed{\text{Childhood tumor}}$

Site – Posterior fossa (cerebellum)

Extremely radiosensitive

Spreads via CSF & produces DROP METASTASIS in Cauda equina

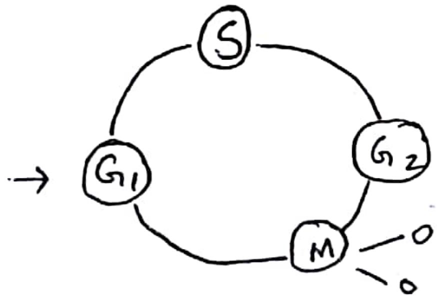
Ependymoma

Arises from ependymal lg lining of ventricles.

Small blue round cell tumors

1. Neuroblastoma
2. Retinoblastoma
3. Medulloblastoma
4. Embryonal rhabdomyosarcoma
5. Lymphoma (Lymphoblastic)

Normal cell cycle regulation.



Orderly progression of cell through cell cycle is brought about by cyclins & cyclin dependent kinases (CDKs) [Protooncogenes]

Cyclins	CDKs	
Cyclin D	4, 6	G_1 to S transition
cyclin E	2	
cyclin A	2	G_2 to M transition
cyclin B	1	

G_1 S transition — cyclin D-CDK4 Most imp.; cyclin E-CDK2

G_2M t — cyclin A/CDK2; cyclin B/CDK1

cyclin A/CDK$_2$ → Takes the cell through G_2 phase upto prophase of mitosis

cyclin B/CDK$_1$ → Regulates all initial events of mitosis beyond prophase.

Inhibitors of cyclins/CDKs

↳ Inactivate cyclins/CDKs and stop the cell in the cell cycle
- G_1 arrest
- G_2 arrest.

CIP/KIP Family (CDKN1A-D)

$\boxed{p21}$ → Induced by p53

$\boxed{p27}$ → Induced by TGFβ

p57

Non Specific

Ink4 family/(ARF) (CDKN2A-C)

p14, p15, 16, 18 19

Specifically inhibit cyclin D/CDK

p16 - most imp.

p14 acts by ↑ing p53 by inhibiting MDM2 activity.

Cell Cycle Checkpoint

G1 S Check point

Sensors → ATM, RAD family

Transducers → CHEK kinase

Effectors → p53 dependent mechanism

G2M

Sensors → ATM RAD family

Transducers → CHEK kinase

Effectors
- p53 dependent
- p53 independent

Rb gene

- Tumor supressor gene - on chromosome 13q14
- Also called as Governer of cell cycle
  or
  Molecular on-off switch of cell cycle.
- Rb causes G_1 arrest of cell cycle.

Rb is located on 13q14

↓

Produces Rb protein

Active form (NP)	Inactive form (P)
- Inhibits cell proliferation	- Allows cell proliferation
- Called under/hypo phosphorylated RB	- Called hyperphosphorylated RB.

⇒ Under phosphorylated RB has pockets in which it hides E2F/DP1 transcription factors - These factors are used in S phase for DNA synthesis. Thus causes G1 arrest of cell

⇒ Under phosphorylated RB also Recruits enzyme Histone deacetylase causing compaction of nuclear chromatin thus G1 arrest.

When the cell has to move from G_1 to S phase, cyclin D/CDK4 phosphorylates the underphosphorylated Rb to hyperphosphorylated Rb

↓

releases E2F/GP1 TF from pockets

↓

Cell will proceed to S phase.

$$G_1 \xrightarrow{\text{Rb (hypophosp.)} \ \ominus \qquad \text{Rb (hyperphosp.)} \ \oplus} S$$

Loss of Rb gene produces
- Retinoblastomas
- Osteogenic sarcomas.

Regulatory genes.

4 classes of regulatory genes.

1. Proto oncogenes
2. Tumor suppressor genes/Anti oncogenes
3. Genes for apoptosis
4. Genes for DNA repair.

(1) Protooncogenes :- Cause cell proliferation
Controlled cell proliferation

Protooncogenes can be GF, GFR, STP (Signal Tranduction) NTP (Nuclear Transduction Proteins) cyclins & CDKs

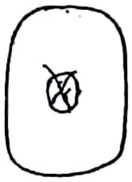
(2) Tumor suppressor gene / Antioncogene
Inhibit cell proliferation by inhibiting

(3) Genes for Apoptosis

- Antiapoptotic
  - BCL2, BCLXL
  - MCL1
- Proapoptotic
  - BAX, BAN
- Arbiters
  - BIM, BID, BAD
  - PUMA, NOXA

(4) Gene for DNA repair.

Molecular basis of Cancer



$\xrightarrow[\text{damage}]{\text{Regulatory gene}}$ Undergoes 30 population doublings. from a single ~~go~~ transformed cell.

↓

Kinetics of tumor -cell growth

1 gm cancer $\bar{c}$ 10^9 cells is formed

↓ 10 population doublings

1 kg Cancer cells $\bar{c}$ 10^{12} cells is formed (Largest Ca mass compatible $\bar{c}$ life)

Genetic damage lies at the heart of carcinogenesis

Regulatory genes are damaged, tranformed cell enters cell cycle & under goes population doublings leading to cancer.

↓

Tumor cells establish their own blood supply by producing — VEGF, bEGF, PDGF

Tumor angiogenesis

↓

Local invasion & distant metastasis

[Phenotypic attributes of Ca cell & are acquired in step wise fashion]

Mechanism of Metastasis

(1) Metastatic sub clone is formed, which shows decreased expression of E cadherin → loosens up from the main tumor.

(2) Metastatic subclone produces enzymes that degrade BM & extracellular connective tissue

e.g Type IV colagenase
Matrix metalloproteins
Plasminogen activator.

(3) Metastatic subclone shows expression of laminin & fibronectin receptors by which they attach to laminin & fibronectin in the connective tissue.

(4) Some cancers also produce autocrine motility factors which help in ca metastasis.

(5) Cancer cells enter the blood vessels where they attach to

⑥ Tumor emboli gets out of blood vessels at a distant site & produces metastatic deposits.

Epithelial to mesenchymal transition (EMT)

Cancer cells acquire a mesenchymal phenotype (e.g; Spindle shape) for metastasis

2 genes for EMT → SNAIL, TWIST } ↓ Ecadherin.

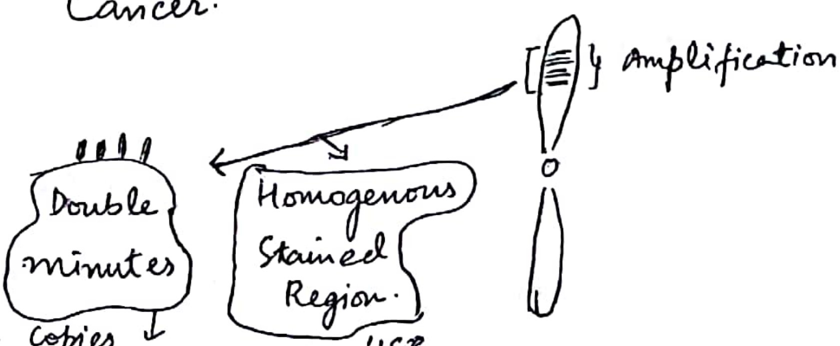
Regulatory genes.

① Proto oncogene (Controlled cell proliferation)

Activated Protooncogene
↓
Oncogenes
↓
Produce onco proteins
↓
Cancer.

- → Translocation
- → Point mutation
- → Amplification
- → Over expression.

POTA



multiple copies separated from chromosome & form small chromosome like structure.

(I) Growth factors

SIS (PDGF-β) $\xrightarrow{\text{Overexpression}}$ Astrocytoma

HST 1 (FGF) $\xrightarrow{\text{Overexpression}}$ Osteosarcoma

INT-2 (FGF3) $\xrightarrow{\text{Amplification}}$ Bladder Ca, Stomach Ca

HGF $\xrightarrow{\text{Overexpression}}$ HCC, Thyroid Ca

(II) GFR (Growth Factor Receptors)

EGFR (Epidermal Growth Factor Receptor)

- ERB-B1 $\xrightarrow{\text{Over expression}}$ Adeno Ca Lung
- ERB-B2 (Her-2-neu) $\xrightarrow{\text{Amplification}}$ Breast Ca, Ovarian Ca

RET $\xrightarrow[\text{(Activation)}]{\text{Point Mutation}}$ - MEN 2A, MEN 2B, Medullary Ca Thyroid.

RET $\xrightarrow[\text{(Inactivation)}]{\text{Point Mutation}}$ ~~Hersh Hershprung disease~~ HIRSCHSPRUNG Disease [Congenital Mega Colon]

KIT (CD 117) $\xrightarrow{\text{Point Mutation}}$ GIST, Soft tissue tumors

ALK $\xrightarrow{\text{Translocation}}$ Adeno ca lung, Lymphoma.

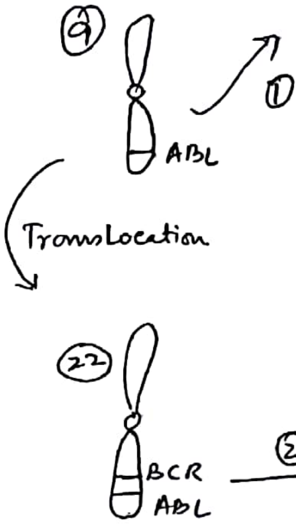
III Signal transduction proteins

ⓐ ABL $\xrightarrow{t(9:22)}$ CML
ALL
AML (Latest)

ABL encodes for a protein with tyrosine kinase activity

↓

Causes signal transduction through Myeloid & lymphoid cells.



→ BCR ABL Fusion gene

↓

Fusion protein $\bar{c}$ very high tyrosine kinase activity.

Myeloid cells	Lymphoid cells
↓	↓
210 KD size	190 KD size
↓	↓
CML	ALL

ⓑ RAS

→ Most common Oncogenic mutations in human Cancers → RAS Mutations.

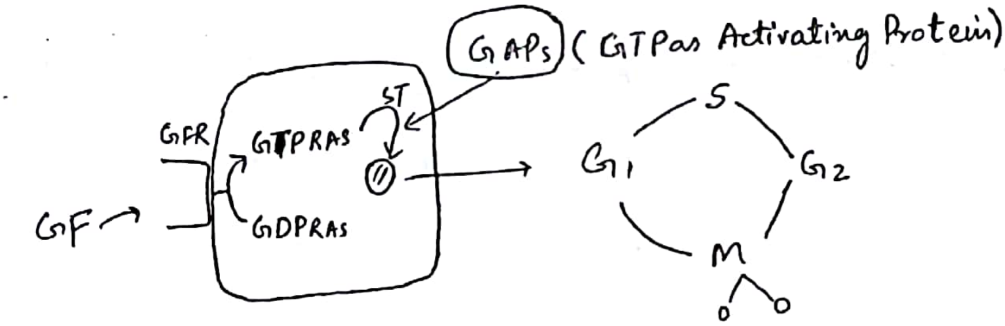
K RAS $\xrightarrow{PM}$ Colon, lung, pancreatic Ca

H RAS $\xrightarrow{PM}$ Urinary bladder & kidney tumor.

N RAS $\xrightarrow{PM}$ Melanomas & hematopoietic tumors.

2 forms of RAS protein

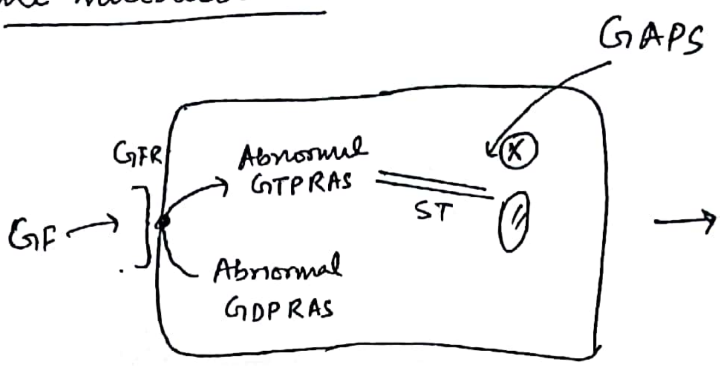
Active	Inactive
GTP RAS	GDP RAS
Allows cell proliferation	Prevents cell proliferation



GF + GFR → GDPRAS is converted to GTPRAS
↓
Causes ST
↓
Cell proliferation

After limited cycles
↓
GAPs → Pull the Phosphate group from GTP RAS & Converts in GDP RAS
↓
Stops ST → Stops cell proliferation

Point mutated RAS



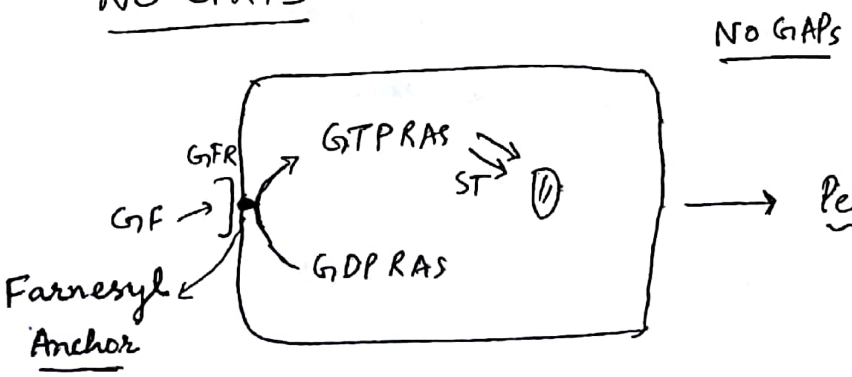
→ Cell proliferation Continues

Due to point mutation in RAS, GAPs are unable to pull 'P' group from abnormal GTP RAS.

↓

RAS Remains in persistant Signal Transduction state & produces tumors.

No GAPs



→ Persistant ST State

GAPs are produced by NF 1 gene

↓

loss of which leads to Persistant ST state & produces Tumors.

NF 1 Syndrome

(c) BRAF

BRAF (V600) $\xrightarrow{PM}$
- Hair cell Leukemia
- LCH
- Papillary Ca Thyroid
- Astrocytoma
- Colonic Ca.

HPLC A

Drug – Vemorafinib

(d) β Catenin

β Catenin $\xrightarrow{PM}$ HCC, Hepatoblastoma.

(e) NOTCH

NOTCH $\xrightarrow{PM}$ Leukemias, Lymphomas.

(IV) Nuclear TransCription Proteins

C MYC $\xrightarrow{t(8;14)}$ Burkits Lymphomas

N MYC $\xrightarrow{Amplification}$ Neuroblastoma.

(V) Cyclin / CDKs

Cyclin D_1 $\xrightarrow{t(11:14)}$ Mantle Cell Lymphoma.

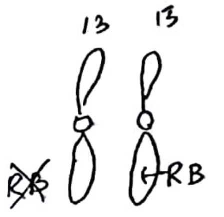
Cyclin E $\xrightarrow{\text{Overexpression}}$ Breast Ca

CDK 4 $\xrightarrow[\text{amplification}]{\text{PM}}$ Glioblastoma
Melanoma

(2) Tumor suppressor genes

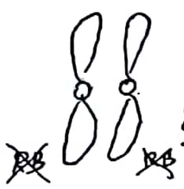
Inhibits cell proliferation

Loss of both copies (Inactivation) of TSG produces Ca.



1st hit

(Heterozygous) - no tumor



2nd Hit

Loss of Heterozygosity
(LOH)

1st tumor supressor gene is RB

Loss — Retinoblastoma
— Osteosarcoma

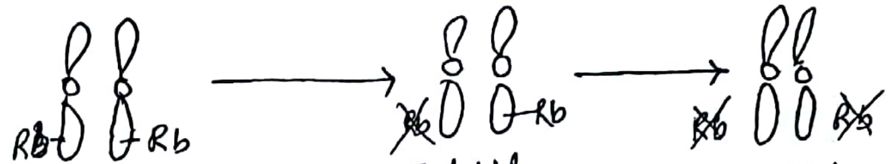
Knudson's TWO HIT HYPOTHESIS for Heriditary Cases (Retinoblastoma) ⇒

Heterozygous → New born

$\xrightarrow{\text{Time}}$

2nd Hit in Retinal cells.

Sporadic Cases



Normal at birth → Ist hit in Retinal cells of eye → 2nd hit in Retinal cells of eye

↓

LOH

↓

Retinoblastoma.

(I) Cell surface

	Function		
TGF-β receptor	Growth inhibition	$\xrightarrow{\text{Inactivation}}$	Ca colon
E cadherin	Cell adhesion	$\xrightarrow{\text{Loss}}$	Ca Stomach

(II) Under the Plasma Membrane

NF-1 (Neurofibromin) or (GAPs) → Inhibits RAS Signal Transduction → NF 1 Syndrome or (Von Reckling hausen's disease)

GAPs are produced by NF-1

↳ Multiple Neurofibromas

↳ Cafe au Lait spots

↳ Lish nodules in Iris (hamartomas)

↳ ↑ risk of brain tumors (optic Nerve glioma)

↳ Juvenile myelomonocytic Leukemia (JMML)

(II) Cytoskeleton

NF2 gene (Chr 22) → Cytoskeletal Stability $\xrightarrow{\text{Loss}}$ NF2 Syndrome

MERLIN is produced by NF2 gene → Cytoskeletal Stability

NF2 Syndrome:
B/L Acoustic schwanoma syndrome
Meningiomas.

(III) Cytosol

APC gene (Chr-5) → Inhibits ST by WNT pathway → Familial Adenomatous Polyposis coli Syndrome (FAP synd.)

APC gene ↓ APC protein → Inhibits ST by WNT pathway

- Multiple adenomatous polyps in colon
- Minimum 100 polyps needed for diagnosis.

Polyps appear by teenage
↓
if untreated
↓
Risk of progression to Ca colon is 100%

(IV) PTEN (Chr-10) → Inhibits ST by PI-3 kinase pathway → Endometrial & prostate ca.

(V) Nucleus

Rb → Retinoblastoma
→ Osteogenic sarcoma

p53 (Chr. 17) → Li Fraumeni syndrome

M/c gene mutated in human Ca → p53 gene.

WT-1, WT-2 } Chr. 11 → Wilms Tumor

p16 → Malignant Melanoma

ARF/INK4A

BRCA-1 (Chr. 17) → Herididitary breast & ovarian Ca (females)
Prostate Ca

BRCA-2 (Chr 13) → Heriditary breast & ovarian Ca (females) Prostate Ca.
→ Heriditary male breast Ca

(3) Genes for Apoptosis

BCL2 $\xrightarrow{t(14:18)}$ Follicular lymphoma.

Chr 14 - IgH gene
Chr 18 - BCL 2 gene

(4) Genes for DNA repair

DNA repair defect syndrome

- Lynch / HNPCC syndrome] A Dominant
- Xeroderma pigmentosa
- Ataxia Telangectasia
- Bloom's syndrome
- Fanconi's Anemia

(Xeroderma pigmentosa, Ataxia Telangectasia, Bloom's syndrome, Fanconi's Anemia) } A Recessive

Genes for DNA repair are of 3 categories

(I) Mismatch repair genes

Act as spell checkers when a strand of DNA is replicating

Loss of mismatch repair genes → Spelling mistakes accumulate in new strand DNA

the cell which gets this DNA is said to Have RER phenotype (Replication ERROR)

Spelling mistakes also produce microsatellite instability.

<u>Micro Satellites</u> → Tandem repeats of 1-6 nucleotides scattered throughout our genome.

Fixed for a person & fixed for life.

Also called as Molecular fingerprints.

<u>Loss of mismatch repair gene is associated with Lynch Syndrome</u>

(↑ risk of developing colonic Ca)

② <u>Nucleotide Excision Repair Gene</u> (NER)

NER genes remove UV light induced pyrimidine dimers from DNA.

Loss → Xeroderma pigmentosa
- Photosensitivity
- 200 times of ↑ed risk of developing Cutaneous Ca
  - SCC
  - BCC &
  - M. Melanoma

T≡T <u>dimers</u>

③ Genes for Repair by Homologous Recombination

Repair double Stranded DNA breaks which Can be produced by ionizing radiation.

ⓐ ATM gene – Sensor of DNA Damage

Loss produces Ataxia talangectasia

Cerebellar Ataxia

Oculo cutaneous talengectasia

Def. of Ig A antibody

↓

(Repeated infection) cause of death

↑ed risk of developing Lympho reticular Tumors.

ⓑ Gene for enzyme BLM helicase

Loss → Bloom's Syndrome

growth retarelation

mental retardation

↑ed risk of developing lympho reticular tumors.

ⓒ Fanconi Anemia gene

Loss → Fanconi Anemia

Heriditary aplastic Anemia

Aplasia of radius & thumb bone

Hypoplastic kidney & Spleen.

8 Hallmarks of Cancer & 2 enabling factors.

8 Hallmarks

① Self sufficiency of Growth signals.

② Insensitivity to growth inhibitory signals.

③ Altered cellular metabolism

Warbug effect Also called as Aerobic glycolysis

It provides rapidly dividing Ca cells with metabolic intermediates that are needed for synthesis of cellular components.

Mitochondria manufactures
↓
other building blocks of Ca. cells

④ Evasion of apoptosis

⑤ Limit less replication potential - due to reactivation of telomerase

⑥ Sustained angiogenesis

⑦ Ability to invade and metastasize

⑧ Ability to evade host immune response.

2 enabling factors

→ Genomic instability

→ Tumor promoting inflammation.

Carcinogens

Chemicals

Arsenic → Lung & Skin

Asbestos → Lung Ca, Mesothelioma
Oesoph., gastric, & colonic Ca

Benzene → AML

Beryllium → Lung Ca

Cadmium & its compounds → Prostate Ca.

Nickel & Chromium → Angiosarcoma, liver

Azo dyes → HCC

β Naphthylamine → Bladder Ca

Nitrosamine & nitrites → Oesophageal & gastric Ca

Aflatoxins → HCC

Carcinogens → Initiators → causes DNA damage
→ Promoters → stimulate genetically damaged cells to proliferate.

e.g Promoters
Hormones like Estrogen, DES (diethyl Stilboestrol)
Saccharin
Phenol
Phorbol esters.

Complete Carcinogens
↳ Capable of both Initiation & promotion.

Carcinogens
→ Direct acting
→ Indirect acting.

Direct acting
e.g alkylating agents
They require no metabolic conversion to become active carcinogen

Indirect acting.
e.g aromatic amines
benzo pyrines
They require metabolic conversion to become ultimate Carcinogens.

Radiation

UV light

UVB Associated with cutaneous Ca.

Xeroderma pigmentosa are at ↑ risk of developing cut. Ca
- SCC
- BCC
- & M. Melanoma

Ionizing Radiation

All leukemia except CLL (MC)

Papillary thyroid Ca

Ca breast, Ca lung.

Ca Salivary gland (Muco epidermoid Ca).

Biological Carcinogens.

Viruses

DNA
- HPV
- EBV
- HBV
- HHV8

RNA
- HTLV-1
- HCV

Bacteria

H. pylori
- MALToma
- gastric Ca

(1) HPV

70 serotypes are known

3 groups

- Low risk serotypes — 6 & 11 ⇒ (condyloma accuminatum) Veneral warts
- Intermediate serotypes — 31, 33
- High risk serotypes — 16, 18

Intermediate & High risk serotypes → Cancer

- Ca Cervix
  Ca Vulva
  Ca Vagina
- Ca Anal Canal
- Ca Penis
  Ca Larynx

Viral DNA encodes for 2 viral proteins

E6
- Inactivates p53 protein
- ↑ TERT + ↑ telomerase expression

E7
- → Inactivates Rb protein
- → Inactivates p21 & p27.

(2) EBV

DNA virus

Infectious Mononucleosis
Oral Hairy Leukoplakia in HIV(+)
] Benign Reactive.

Tumors

Nasopharyngeal Ca
Burkittes lymphoma
Hodgkins lymphoma
Bcell lymphomas in immunocompromised.
Extranodal NK T cell lymphoma.

Produces 2 ~~produce~~ proteins responsible for excessive cell proliferation

→ LMP-1
→ EBNA 2

(3) HBV

→ HCC

- Repeated cycles of injury and regeneration lead to accumulation of mutation.
- Some viruses have $\boxed{\text{HBx gene}}$
  ↓
  Interferes with p53
  ↓
  Ca

④ HHV8 [Kaposi's sarcoma Herpes virus 8]

Causes → Kaposi's Sarcoma
→ Primary effusion lymphoma (variant of DLBCL)

RNA Virus

① HTLV-1

It Causes
Arthritis
Uveitis
Tropical spastic Para paresis

Tumor – Adult Tcell Leukemia/lymphoma.

HTLV-1 codes to TAX protein
↓
Causes uncontrolled cell proliferation.

② HCV
[HCC
Spleenic marginal Zone lymphoma

Paraneoplastic syndrome

(1) Endocrinopathies

(a) Hypercalcemia (MC)

Due to production of PTH related protein. by tumor cells.

Tumors – SCC (Lung, Skin), Ca breast, RCC, Adult T cell leukemia/lymphoma.

(b) Cushing syndrome

Due to ACTH production

Tumor – Small cell Ca lung.

(c) SIADH

Due to ADH production

Tumor – Small cell Ca lung.

(d) Hypoglycemia

Due to production of Insulin or Insulin like substances.

Tumor –
- Ovarian Ca
- Fibrosarcoma
- HCC

(e) Carcinoid Syndrome

Due to production of Bradykinin & serotonin

Tumors –
- Bronchial Carcinoids
- HCC

(f) Polycythemia
Due to erythropoietin

Tumors — RCC
HCC
Cerebral Hemangioma.

② Nerve & muscle

Myasthenia gravis — Ca lung
Thymoma.

Immunologic in origin

③ Cerebral degeneration

Ca lung
Hodgkins lymphoma

④ Skin
Acanthosis Nigricans.
Due to production of epidermal growth factor.

Tumor — Gastric Ca
Lung Ca

⑤ Hypertrophic osteoarthropathy – Ca lung.

(5) Trousseau syndrome
(Migratory Thrombophelibitis)

Tumor cells activate clotting

- Pancreatic Ca
- Lung Ca

(6) Marantic endocarditis/Non bacterial Thrombotic endocarditis

In advanced Malignancies

Tumor Markers:

(1) Hormones

(a) Calcitonin – Medullary Ca thyroid.

(b) Catecholamine – Pheochromocytoma

(c) β HCG – Trophoblastic tumors of Chorio Carcinoma.

(2) Oncofetal antigens

(a) AFP (Alpha Feto Protein) – HCC, Hepatoblastoma
Yolk sac tumor
(No seminomatous germ cell tumor)

(b) CEA (Carcino Embryonic Antigens)
→ Ca colon, Pancreas, lung, stomach

③ Specific Proteins

(a) Immunoglobulins — Multiple Myeloma.

(b) PSA — Prostate Ca.

④ Iso-enzymes

(a) PAP (Prostate Acid Phosphatase) — Prostate Ca

(b) Neuron specific Enolase — Neuroblastoma
Small cell Ca
Neuroendocrine tumors.

⑤ Mucins

CA 125 → Ovarian Ca

CA 19.9 — Colon & prostate Ca.

CA 15.3 — Breast ca

⑥ Cell free DNA markers (LIQUID BIOPSY)

TP53, APC, RAS in stool & serum — Colon ca

TP53, RAS in stool & serum — Pancreatic Ca

TP53, RAS in sputum & serum — Lung Ca

TP53, in urine & serum — Bladder Ca

Tumor Markers detected in Tissue by IHC

1. Carcinoma
   - Cytokinin (m. imp)
   - EMA (Epithelial Membrane Antigen)
   - CEA (Carcinoembryonic Antigen)

2. Lymphoma
   - LCA (Leucocyte common Antigen)

3. Sarcoma – Vimentin
   - Ewing sarcoma – CD99
   - Synovial sarcoma – CD99, BCL2
   - Rhabdomyosarcoma – Desmin, MyoD1
   - Leiomyosarcoma – Smooth Muscle Actin + (SMA)
   - Osteosarcoma
     - Osteopontin
     - Osteonectin
     - Osteocalcin
   - Chondrocytoma – S-100
   - Liposarcoma – S-100

4. Mesothelioma
   - ~~S-100~~
   - Cytokeratin 5/6
   - Mesothelin
   - Calretenin

5. Malignant Melanoma
   - HMB45
   - Melan A
   - S100

6. LCH
   - CD1a
   - S-100
   - Langerin (CD 207)

7. Small cell Ca
   Neuroblastoma
   Neuro endocrine tumor
   - Synaptophysin
   - Chromogranein
   - Neuro Specific enolase (NSE)
   - S-100

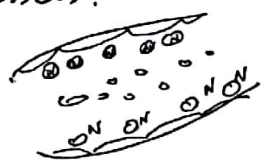
8. Schwan cells
   Neurofibroma
   Schwanoma
   - S-100

9. TTF-1 (Thyroid Transcription Factor)
   - Lung Ca
     - Adeno Ca
     - Small cell ca
   - Thyroid Ca

10. HCC
    - Arginase 3
    - Hep Par 1

I Vascular events

1. Transient vasoconstriction
   ↓
2. Massive vasodilation
   ↓
3. Increased vasular permeability. → exudate formation (Most imp)
   ↓
4. Stasis of cells in Blood vessels.



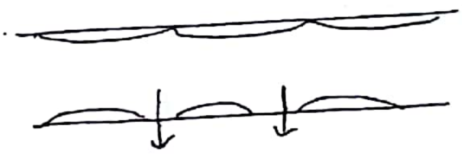
   ↓
5. Leucocyte margination to the periphery.

II Cellular events

- Adhesion & transmigration
- Chemotaxis
- Phagocytosis

Mechanisms of ↑ vascular permeability

1. Endothelial contraction (M/c mech)
(occurs in post capillary venules)



Mediators
- → Histamine
- → Bradykinin
- → Substance P
- → Leukotriene

⇒ Immediate Transient response

② Endothelial retraction / Junctional reorganisation

Delayed type of contraction (venules & capillaries)

Mediators (cytokines) → IL-1
→ TNF α

⇒ ② Delayed, sustained response

③ Direct Injury

(Seen in venules, arterioles, capillaries)

Severe Injury

e.g Severe burns
Chemicals
Toxins

↓

Cells undergo necrosis & detach.

↓

Fluid leakage which starts immediately

↓

Leakage continues till a new cell regenerates

② ⇒ Immediate sustained response

Mild Injury

e.g mild sunburn.

↓

endothelial cells die after a few hours due to apoptosis.

↓

Fluid leakage starts after few hours

↓

Leakage continues till a new cell regenerates

⇒ ① Delayed sustained response

(4) Increased Transcytosis

⇒ Passage of liquid across the channels formed in the endothelial cell cytoplasm

⇒ Channels are formed close to the Junction

Mediator – VEGF

(II) Cellular events

(1) Adhesion and transmigration

Rolling ⟶ loose adhesions ⟶ firm adhesions
↓
Transmigration (Diapedesis)

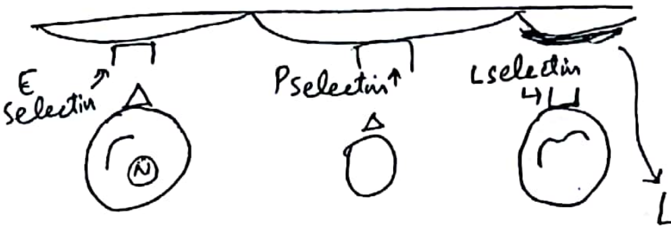
Adhesion molecules ← - - - - - - - - 
↓
Substances that coat the surface of neutrophills and surface of endothelial cells & help them to stick together.

4 Families of adhesion molecules

(I) Selectins – Bring about Rolling & loose adhesions

E selectin	CD 62E	Endothelium
P selectin	CD 62P	Platelets, endothelium
L selectin	CD 62L	Leukocytes

Complementary molecules
↳ Sialyl Lewis X
(Sugar formed by fucose metabolism)
[E & P]



Sialyl Lewis X.
(on the surface of Leukocytes)

↳ Sialyl Lewis X in mucin like ~~glycolipids~~ glycoproteins that coat the endothelium.
(for [L])

Leucocyte Adhesion deficiency type II
(LAD type II)
⇓
deficiency of Sialyl Lewis X due to defect in fucose metabolism.

(II) Immunoglobulin Superfamily

ICAM → ①, ②
(Intercellular Adhesion Molecule)
VCAM → ①, ②
(Vascular Cell Adhesion Molecule)

Both are found on endothelium

(III) Integrins

β_2 integrins
e.g LFA1 / MAC1
(CD11) (CD18)
β_1 integrins → VLA4

Found on leucocytes

Responsible for $\boxed{\text{FIRM ADHESIONS.}}$

IV CD31/PECAM-1

(Platelet Endothelial Cell Adhesion) Molecule

- Transmigration
- Homotypic adhesion molecule
- CD31 is found on leukocyte & enothelium.

Neutrophils produce enzyme type IV collagenase
↓
breakes type IV collagen (Basement Membrane) & comes out of the vessel.

LAD type I

Autosomal recessive disorder

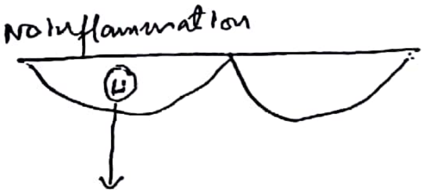
Deficiency of β_2 integrins LFA-1/MAC-1

There is mutation in MAC 1 (CD18) gene

C/F - Recurrent bacterial & fungal infection.
Impaired wound healing.
Delayed umbilical cord separation.
Leukocytosis (↑ TLC).

Mechanism of appearance of Adhesion molecules.

① Redistribution → P selectin



Weibel Palade body in endothelial cell cytoplasm
[contains P selectin)

During inflammation

Mediators → Histamine
→ Thrombin
→ PAF

↓

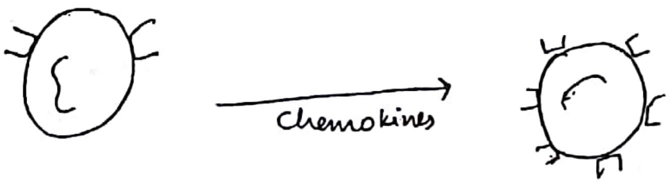
redistribute P selectin to endothelial cell surface within few minutes

(2) Induction → E selectin, ICAM, VCAM
↓
Fresh synthesis during inflammation

Mediators that induce → IL 1, TNFα

Time taken is hours.

(3) Increased avidity of binding
↓
(Strength)
⇓
Integrins



↑ the no. of integrin molecules on leukocytes

↑ the strength of binding of integrins many times.

⇒ Weibel Palade bodies are ultrastructural markers of endothelial cells (EM)

② Chemotaxis

Def. Locomotion orientend along a chemical gradient.

Chemotactic agents

LTB4
C5a
IL8
Bacterial Products
(containing N formyl methionyl residues)

Serpentine Receptor
or
7 transmembrane G coupled receptor.

When these ligands bind to 7 transmembrane G coupled receptors

↓

↑↑ Ca^{2+} in the cytosol

↓

Polymerisation of actin filaments at the leading edge

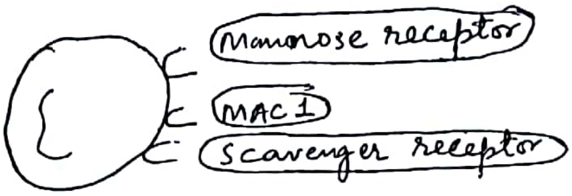
↓

Pseudopod formation

Defects In
DM, Malignancy, severe burns, CRF

③ Phagocytosis

(a) Recognition & attachment.



3 receptors that help the neutrophil to recognize & attach to the bacteria

- Mannose receptor
- MAC 1 integrin (CD18)
- Scavanger receptor

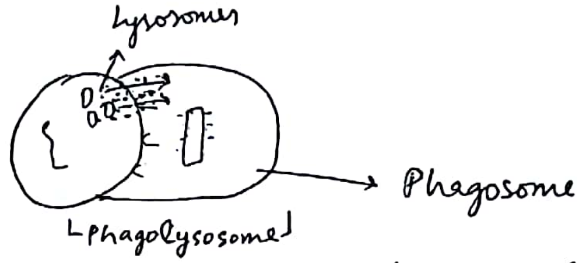
Opsonization

↑es the efficiency of phagocytosis.

Opsonins → Substances that coat the bacteria & make it tasty for neutrophil.

- → C3b, C3bi (C4b, C4bi)
- → IgG antibody
- → Plasma proteins
  - → CRP
  - → Mannose Binding lectin
  - → Fibronectin
  - → Fibrinogen.

(b) Engulfment.



Pseudopods flow around the bacteria & bacteria is enclosed in a phagosome

Phagosome fuses with lysosome to form phagolysosome

- All enzymes are discharged in phagolysosome.

Chediak Higashi syndrome – defect in engulfment

↳ Autosomal recessive disorder

↘ Failure of fusion of phagosome & lysosome.

C/F – Repeated infections

- Oculocutaneous albinism & Silvery grey hair.
- Nerve conduction defects.
- Platelet function defects leading to bleeding.

PBS Neutropenia with giant granules in leukocytes

⇒ Lyst gene mutation are seen.

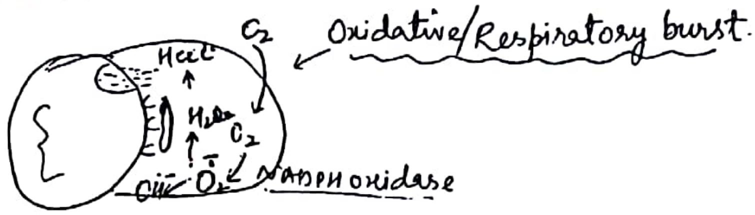
⇓

Absence of DOCKING PROTEIN

Needed for fusion of lysosomal membrane with phagosomal membrane.

(C) Killing → O_2 dependent method (Main method)
→ O_2 independent method

Oxygen dependent method



$H_2O_2 \xrightarrow[Cl]{MPO} \boxed{HOCl^-}$ kills bacteria

→ By lipid peroxidation of bacterial membrane

→ Halogenation (bleaching)

H₂O₂ MPO - Halide system kills bacteria, fungi, & parasites

MPO is a lysosomal enzymes.

Defect → Chronic granulomatous diseases
↓
defect of enzymes NADPH oxidase
→ XR (MC) (75%)
→ AR (25%)

Test for diagnosis Nitroblue tetra zolieum Test. (NBT)

Oxygen independent killing

Lysosomal enzymes kill the bacteria

→ **BPI** Bacterial Permeability Increasing Protein

It is a phospholipase
↓
Breaks phospholipids & bacterial membrane

→ **Lysozyme**

It is a muramidase
↓
Breaks glycopeptide coat of bacteria.

→ **Lactoferrin**

Binds Iron
↓
Iron is unavailable for bacterial growth

⇒ **Major Basic Protein**

Found in eosinophil granules
↓
Toxic to parasites

→ **Defensins**
→ **Cationic proteins.**

} Found in Neutrophils

Mediators of acute inflammation

Cells

Preformed/Primary

- Histamine
- Serotonin
- Lysosomal enzymes

Newly synthesised/Secondary

- Aracadonic Acid mediators
- PAF

Lipid mediators

- Cytokines
- ROS (Reactive Oxygen sp.)
- Chemokines
- Nitric oxide
- Substance P

Plasma

- Kinin cascade
- Complement cascade
- Coagulation cascade
- Fibrinolytic cascade

Preformed / Primary mediators

① **Histamine** First mediator to be produced

Source → Richest source is Mast cell

Others – Basophils, Platelets

Stumili for release

⇒ IgE Ab binding to receptors on mast cells

Post Cap. Venules

⇒ C3a, C5a } Anaphylotoxins

⇒ IL-1, IL-8

⇒ Histamine Releasing proteins.

⇒ Physical agents like heat, cold & trauma.

Actions

① Vasodilation

② ↑ vascular permeability → Causing Immediate Transient response.

③ Vasoconstriction (Large vessels due to muscular layer presence)

④ Bronchospasm.

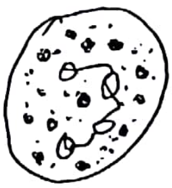
(2) Serotonine (5HT)

Richest source → Platelets
Others → Enterochromaffin cells.

Actions Platelet aggregation
Other actions are same as histamine.

(3) Lysosomal Enzymes

Found in granules of neutrophils



Primary/Azurophilic granules
- Large coarse granules

p ANCA → MPO
→ BPI
→ Acid hydrolases
c ANCA → Neutral proteases
→ Lysozymes
→ Phospholipase A_2
→ Defensin
→ Cationic Proteins

Secondary/specific granules
- Small fine granules

Lactoferin
Alkaline phosphatase
Type IV collagenase
Gelatinase
Lysosyme
Phospholipase A_2
Vit B12 binding proteins

ANCA Antibodies against enzymes found in primary granules of Neutrophils

P ANCA → MPO

C ANCA → Proteinase 3 (Neutral proteases)

Newly synthesised mediators.

① Platelet activating Factor. (Lipid meddator)

Source – All leukocytes & mast cells.

Actions
- Vasodilation
- ↑ Vascular permeability
- Vasospasm
- Platelet aggregation
- Brochospasm
- Chemotactic
- Angiogenesis
- Cell to cell signal transduction.

② A. acid mediators

20 Carbon polyunsaturated Fatty Acid
Found esterified in membrane phospholipids.

Membrane phospholipids
↓ Phospholipase
A.A.

- A.A. $\xrightarrow{Lox}$ LT, Lipoxins
- A.A. $\xrightarrow{Cox}$ PG, TxA_2

$\underbrace{\text{LT, Lipoxins, PG, } TxA_2}_{\text{Eicosanides}}$

Source – All leucocytes, mass cells, endothelial cells.

Steroids inhibit phospholipase

AA.

↓ cyclooxygenase (COX) ← ⊖ COX 1 / COX 2 inhibitors

PGG_2

↓

PGH_2

Endothelium → Prostacyclin synthetase → PGI_2 (Prostacyclin)
- Inflammatory mediator
- Causes vasodilation
- Inhibits platelet aggregation.

PGD_2, PGE_2, PGF_2

Platelets → Thromboxane synthetase → TXA_2 (No role in inflammation
- plays role in hemostasis)
  - → Platelet aggregation
  - → Vasoconstriction.

⇒ Increase vascular permeability

⇒ Cause vasospasm. & Bronchospasm.

⇒ Pain & Fever (PGE_2)

⇒ PGD_2 & PGF_2 ⇒ Chemotactic.

⇒ $PGF_{2\alpha}$ ⇒ Causes uterine smooth muscle contraction.

AA

↓ 5 LOX (neutrophils) ⊖ ← LOX inhibitors
(Lipooxigenase)

5 HPETE ---12 LOX (platelets)---→ Lipoxins

Lipoxins: LXB4, LXA4

Antiflammnetory

They inhibit Neutrophil chemotaxis

Inhibit NK cell activity.

5 HPETE → LT A4

LT A4 → LT B4 → Powerfull Chemotactic agent for Neutrophils

LT A4 → LTC4 → LT D4 → LT E4

Slow reacting substances of anaphylaxis

Action :- ↑ vascular permeability

Cause vasospasm & bronchospasm.

Lipoxins are produced by transcellular biosynthesis

↓

2 Cells are cooperating for production.

(3) Chemokines Belong to family of cytokines

Short chain polypeptides that cause chemotaxis.

4 categories

(I) α chemokines / CXC chemokine

Chemotactic for Neutrophils

e.g: IL 8

(II) β chemokines / C-C chemokines

Chemotactic for all except Neutrophils.

e.g: Eotaxin → only for eosinophils

Rantes → eosinophils + T lymphocytes.

MCP 1 ⟶ Monocytes

(Monocyte chemoattractant Protein 1)

MIP-1α ⟶ Monocytes & Macrophages

(Macrophage Inflammatory Protein)

(III) gamma chemokines / C - Chemokines.

e.g: lymphotactin → Lymphocytes.

(IV) CX_3C Chemokines

Only one member - Fractalkine - chemotactic for monocytes

Chemokine receptors → CXCR4, CCR5 } act as coreceptors for HIV

(3) Cytokines

Macrophage & dendritic cells.

Source

IL-1 }
TNFα] TNFα in addition is also produced by T cells & mast cells.

Action – (I) Systemic acid phase reaction

IL-1 }
TNFα } Fever, Increased sleep,
IL6 → MC. ↓ appetite
↑ TLC
↑ ESR
↑ CRP

TNFα also regulates energy balance by causing lipid and protein mobilization & suppressing appetite

↓

thus causing ↑ TNF levels

⇓

Cachexia

Cancer cachexia → TNFα.

(II) Endothelial activation

↑ expression of endothelial adhesion molecules

↑ production of mediators – cytokines chemokines & AA mediators

↑ procoagulant activity.

(III) Leukocyte activation

TNF ⇒ Microbicidal activity of leukocytes

(IV) Fibroblast activation

IL 1 - Fibroblast proliferation &
Synthesis of collagen.

(5) Nitric Oxide

Arginine $\xrightarrow{\text{NO synthetase}}$ NO

NO synthetase is found in → endothelial cells

Endothelial cells → $\boxed{\text{eNO}}$ → Vasodilation

Macrophages → $\boxed{\text{i NO}}$ (inducible) → Produced during inflammation
- Microbicidal gas.

Neurons in brain → $\boxed{\text{n NO}}$ (neuronal) → Neurotransmitter in Brain.

(6) Substance P (Neuropeptide)

Source - Leukocytes
Sensory Nerves. (CNS & PNS)

Actions
- Pain
- ↑ vascular permeability
- Regulation of BP

PAIN mediators.
- PGE2
- Bradykinin
- Substance P. → M.Imp

Mediators comming from Plasma

(1) Kinin Cascades [generate bradykinin]

$$\text{Prekallikrein} \xrightarrow[\text{(Hegeman factor)}]{\text{XIIa}} \text{Kallikrein}$$

Kallikrein
↓
High M.W kininogen
↓
Bradykinin

Action
- Pain
- ↑ vascular permeability → M. Imp
- Vasodilation
- Vasospasm
- Bronchospasm.

(2) Complement Cascade

[Set of 20 proteins found in plasma]

3 pathways for complement activation

Classical pathway
- Starts c̄ activation of C1
- C1 is activated by Ag-Ab complex.

Lectin Pathway
- Starts c̄ activation of C1.
- C1 is activated by baterial lectins and mannose binding lectins

Alternate Pathway
- Starts c̄ activation of C3.
  No participation of C1, C2, C4
- C_3 is activated by
  - LPS (lipoP.S)
  - Cobra venom
  - Aggregates of IgA antibody.

Mediators produced

C3a, C3bi } Opsonins

C3a, C5a } Anaphylotoxins (cause release of histamine from mast cells).

C5a } Chemotactic for Neutrophils, eosinophils & monocytes

C5-9 } Membrane Attack Complex (MAC)

③ Coagulation Cascade

Mediators

ⓐ Fibrinogen → Opsonin

ⓑ Thrombin → Main ~~lin~~ link b/w inflammation & coagulation.

Causes redistribution of Pselectin.

Induces COX enzymes in endothelial cells.

↑ expression of adhesion molecules on endothelial cells.

ⓒ Fibrinopeptides → Vascular permeability

→ Chemotactic

④ Fibrinolytic Cascade

Plasminogen

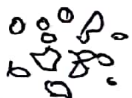
↓ Tissue plasminogen Activator (TPA), Urokinase, Streptokinase

Plasmin

↓

clot

↓



Fibrinogen Degradation Products

FDP → ↑ vascular permeability

Chemotactic

NETs

Neutrophil Extracellular Traps.

Extracellur Fibrillar Network formed by Neutrophills to trap bacteria.

Formed from the nuclear chromatin of Neutrophils

Lysosomal enzymes are discharged in the NETs & kill the bacteria

At the end of NET formation, neutrophils die.

Chronic Inflammation

① Infiltration of tissue by mononuclear cells (monocytes, lymphocytes, & plasma cells)

② Tissue destruction.

Macrophages

⇒ Main cells of chronic inflammation

⇒ Derived from blood monocytes which are produced in bone marrow.

* Resident macrophages eg kupffer cells in liver, microglial cells in brain are derived from stem cells in yolk sac & life span is in years.

⇒ Also called as Histocytes

Liver → Kupffer cells

Brain → Microglial cells

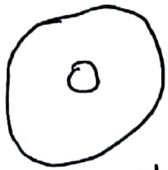
Bone → Osteoclasts

Spleen → Littoral cells
Spleen → Sinus histiocytes

LN → Sinus histiocytes

Lungs → Alveolar macrophages

In chronic inflammation macrophages are activated to kill the bacteria.



Macrophage

- IF gamma
- Uric acid crystals
- Bacterial endotoxins

→ Classical Pathway

M1 macrophage

→ ROS, NO, Lysosomal enzymes

will kill the affecting agent and cause inflammation and tissue destruction.

Cytokines except IF gamma

IL 4
IL 13

→ Alternate Pathway

↓

M2 Macrophage

↓

IL-10
TGF β

→ Tissue repair & fibrosis
→ Antiinflammatory

Fibrosis - TGF β

Antiinflammatory - IL 10
TGF β
IL 6

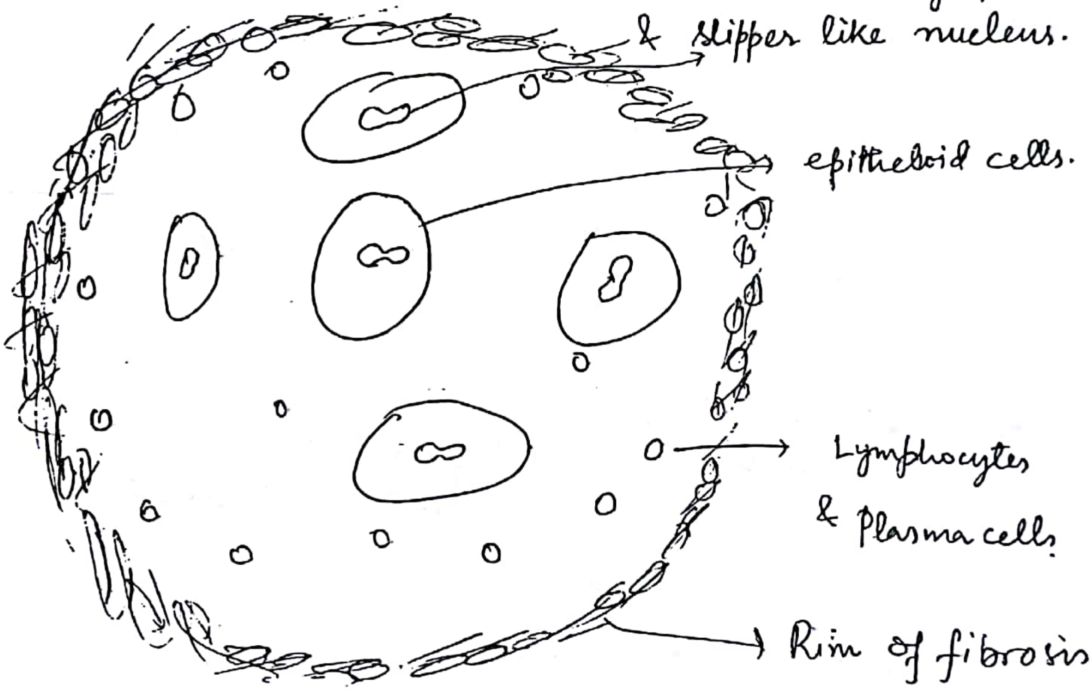
✓ ⇒ IL 6 is both Pro inflammatory & Antiinflammatory

Chronic granulomatous inflammation

Special type of chronic inflammation characterised by granuloma formation.

→ Collection of specialized macrophages called epithelioid cells

↳ Have abundant cytoplasm & slipper like nucleus.



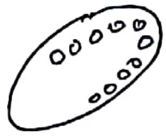
2 types
→ Immune granuloma
→ Foreign body granuloma.

(1). Immune granuloma ~~Found~~ Formed in type IV HR.

→ IFN gamma & TNF α plays an important role.

Causes
- TB
- Sarcoidosis
- Leprosy
- LGV
- Cat scratch disease
- Syphilis
- Crohn's disease
- Hodgkin lymphoma

Immune granulomas may show gaintcells
e.g Langhan's gaint cell.



Multiple nuclei arranged in C or horse shoe shap

⇒ Caseating Granuloma
Caseous necrosis in centre

TB
Syphilis
Fungal infection < Histoplasmosis, Coccidoidomycosis

⇒ Non Caseating

TB
Sarcoidosis (naked granulomas)
↳ No lymphoid & PC surrounding

Hodgkin's lymphoma
Leprosy (tuberculoid)

⇒ Stellate granulomas < Star shapped granuloma, Neutrophilic granuloma
LGV
Cat scratch disease

⇒ Necrotising granuloma
Wegner's granulomatosis
(Small vasculitis)

→ Eosinophilic granulomas
Churg Strauss
(small vessel vasculitis)

→ Dureck granulomas
Cerebral malaria.

② Foreign body granuloma
Formed around a foreign body eg: talc, suture material, dead parasites, uric acid crystals.

↳ Contains foreign body giant cell (numerous)

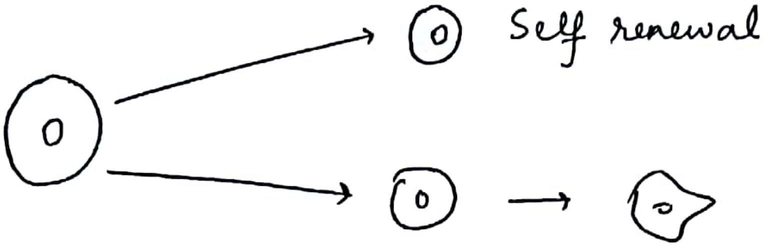
Multiple nuclei arranged in haphazard way
May contain foreign body

Cytokines for Fever
1. IL1 Most imp.
2. IL6
3. TNF α
4. PG E2
5. Ciliary Neurotrophic factor

Cytokines for Septic Shock
- TNF α

Fibrosis - TGF β

Stem Cells



Capacity to generate differentiated cells by asymmetrical replication

2 types

(1) Embryonic stem cells – Isolated from blastocyst

Totipotent - can generate all tissues of body.

(2) Adult or Somatic stem cells

Found in Adult/Normal tissues in special microenvironments called Niches

Can be Pluripotent/Multipotent/Bipotent.

(a) Bone marrow

→ Hematopoietic stem cells

- Pluripotent - give rise to all blood cell lineages.
- Can be obtained from
  - (I) Bone marrow
  - (II) Peripheral blood after injecting GM-CSF
  - (III) Umbilical cord blood

→ Marrow Stromal stem cells

- Mesenchymal S.C.
  Pluripotent
  give rise to adipocytes, endothelial cells, Osteoblasts, chondrocytes

Mesenchymal stem cells are also found in abdominal fat.

b) Liver

Called "Oval cells"

Found in Canals of Hering.

Bipotent
- Hepatocytes
- Bilary cells.

c) Brain

Called Neuronal S.C.

Multipotent.
- → Oligodendrocytes.
- → Neurons
- → Astrocytes

Location
- → Subventricular zones
- → Dentate gyrus of hippocampus.

d) Skin

Found in adnexa
- → Hair follicle bulge
- → Sebaceous glands

e) Limbus of cornea

f) Crypts of Intestine

g) Satellite cells - Sc associated with Cardiac & skeletal muscles.

Use of Stem cells

Used to repopulate the damaged tissue

Wound Healing

Regeneration	Repair
Dead cells are replaced by same cells	Dead cells are replaced by fibrous connective tissue.

① Labile cells / Actively dividing cells

They are in G1 phase of cell cycle.

eg epithelia ⇒ Skin. GIT, Resp. tract urogenital tract.

Stem cells

Hematopoietic cells

Cancer cells

② Stable cells
In G0 phase of cell cycle
They have low replicative potential.
e.g; i) Parenchymal cells of organ
Hepatocytes
PCT, DCT of kidney
Adrenocortical cells.

(ii) Mesenchymal cells
Adipocytes
Osteoblasts
Chondro cytes
Smooth muscle cell
Endothelial cells.

③ Permanent/Non dividing cells
They have left the cell cycle.
They cannot divide at all.
e.g: - Neuronal
- Skeletal muscle
- Cardial muscle

Repair occurs by formation of granulation tissue
Pink, moist & has granular appearance.
M/c → Chronic inflammatory cells/Macrophages
lympocytes, plasma cells.
→ New blood vessels
→ Fibroblasts that synthesize collagen.

Wound healing by Primary intention

Seen in clean, surgical wounds where the edges can be approximated

0 hours → Incision is filled c̄ blood clot.

c̄in 24 hours
- → Neutrophils from margins infiltrates the clot.
- → Mitosis begins in the basal layer of epidermis.

24 - 48 hours → Contineous thin layer of epithelium is formed below the scale/scab.

Day 3
- → Neutrophils are replaced by macrophages
- → Granulation tissue appears.
- → Collagen fibers are evident at the margins of the incision.

Day 5
- → Abundant granular tissue*
- → Neovascularization is maximum.
- → Collagen fibers bridge the incision (i.e they lay down longitudinally)
- → Epidesmis regains full thickness c̄ surface keratinization.

2nd week → ↓ edema, ↓ inflammation
↓ vascularization
→ Proliferation of fibroblasts and accumulation of collagen.
↳ Blanching begins due to ↑ collagen deposition & regression of vascularity

3rd week → Scar is formed $\bar{c}$ maximum fibrosis.

Wound Strength

1st week → 10% of normal unwounded skin.

3rd month → Maximum wound strength
70 – 80% of unwounded skin.
(100% wound strength is never regained).

→ Wound contraction is absent in primary intention healing.

Healing by secondary intention.

Seen in large wounds where wound edges cannot be approximated.

Large amount of granulation tissue is formed

Large scar is formed.

↓

Scar reduces in size – this is called as wound contraction

Brought about by Myofibroblasts.

⇒ Wound contraction is seen in healing by Secondary intention.

Defects in wound healing.

Hypertropic scar :- Raised scar produced due to accumulation of excessive amount of collagen.

⇒ Seen in thermal / traumatic injury.

⇒ Grows rapidly & regresses over several months.

Keloids :- Scar tissue grows beyond the boundaries of the original wound.

⇒ Do not regress

⇒ Genetic predisposition

Exuberent granulation tissue (Proud flesh)

Formation of excessive amount of granulation tissue which protrudes above the level of surrounding skin and blocks the ~~re~~ epithelialization.

Removed by cautery or surgical excision.

Desmoid / ~~Fibroblast~~ Fibromatosis

Excessive proliferation of fibroblasts and other connective elements on site of injury or surgical scar.

Contrature

Exaggeration of wound contraction produces contracture

Common after severe burns

Site – Palm, sole, anterior thorax

Factors that impair wound healing.

1. Infection
2. DM
3. Poor nutritional status < Zn def., Vit C, PEM
4. Mech. factors < Local, Torsion
5. Poor perfusion due to < atherosclerosis, DM, impaired venous drainage
6. Foreign body
7. Type of injury & site of injury

Positive acute phase proteins

Also called as Acute Phase Reactants.

Production increases during inflammation.

CRP
Fibrinogen
Mannose binding Lectin
} Opsonins

SAA [Serum Amyloid Associated]
Ceruloplasmin
Heptoglobin
} Inhibits the growth of microbes.

Prothrombin
Factor VIII
vWF
} Bring about coagulation ~~cases~~

Negative acute phase proteins

Production by liver decreases during Inflammation.

Albumin
Transferrin
Transthyretin
Trans cortin
Retinol binding proteins.

Collagen

⇒ Triple Helix — α_1, α_2, α_3, α_4, α_5

→ any three make the Helix.

Vitamin C is essential for crosslinking of collagen fibres.

Type I collagen

Most abundent

Has high tensile strength

Found in skin, bones, tendons, internal organs & blood vessels.

Type II collagen

Cartilage & Vitreous humor

Type III collagen

Granulation tissue

Embryonic tissue

Uterus

Keloid

Type IV Collagen

Basement membrane.

Composition of basement membrane

Laminin

Type IV collagen

Fibronectin

Proteoglycans.

Amyloidosis

Group of diseases that have in common - deposition of abnormal proteinaceous substance extracellularly.

H&E → Pink homogenous appearance

Physical Nature

Q 1) Electron Microscopy

- Long non branching fibrils
- Indefinite length fibrils.
- 7.5 – 10 nm diameter.

Q 2) X ray crystallography & infrared spectroscopy ⇒ β pleated sheet conformation

Chemical Nature

→ Fibrillar protein
Constitutes 95% of amyloid
15 different types of Fibrillar proteins discovered.

→ P-component Glycoprotein
Constitutes 5% of amyloid.
⇒ PAS +ve & diastase resistant.

Classification

Localized amyloidosis

Systemic amyloidosis
Multisystemic disorder.

Localized Amyloidosis

1) Medullary carcinoma thyroid. — A Cal
A → Amyloid, Cal → Calcitonin.

2) Ahlzimer's disease — A β

3) Isolated atrial amyloidosis — A ANF
ANF → Atrial Natriuretic Factor

4) Type II DM
↳ Amyloidosis in pancreas — A IAPP
IAPP → Islet Associated Pancreatic Peptide

5) Prion disease — Misfolded prion particles.

Systemic / Generalized Amyloidosis

① **Primary amyloidosis** seen in patients of Multiple myeloma & other plasma cell tumors.

AL (Light chains) type of amyloidosis.

λ light chains are more prone to settle down as amyloid

Site - Heart, Kidney, GIT, etc.

② **Secondary amyloidosis** Also called reactive systemic amyloidosis

→ SAA (Serum Amyloid Associated) $\xrightarrow[\text{proteolysis}]{\text{Incomplete}}$ A A type amyloid

Site - kidney, liver, spleen, lungs, etc

M/C organ involved in amyloidosis ⇒ KIDNEY

Causes
- → Older days - TB, DM, Lung abscess, Bronchi abscess.
- Now → Ankylosing spondylitis
  - R A
  - Ulcerative colitis
  - RCC

(3) Hemodialysis Associated Amyloidosis.

Seen in patients who are on long term hemodiplysis for chronic Renal Failure

β_2 microglobulin → $A\beta_2$

Sites - Joints, tendons, synovium

↳ Carpel tunnel syndrome.

(4) Senile Amyloidosis

Seen in old age

Site - Heart liver, spleen, etc

Transthyritin (TTR) is deposited as amyloid → ATTR

↳ (N) serum protein that transports thyroxine & retinol.

(5) Familial Amyloidosis

Familial Amyloidotic Polyneuropathy

- ⇒ ATTR derived from TTR is deposited as amyloid
- ⇒ Site < Sensory Nerves, Autonomic Nerves
- ⇒ Mutated Transthyritin is deposited as amyloid
- ⇒ AD. disorder

Familial Mediteranean Fever

- ⇒ AR disorder. (Recessive)
- ⇒ AA derived from SAA is deposited as amyloid in many organs.
  Fever, effusions seen
- * Pyrin gene mutations are seen

Diagnosis

- FNAC
  - Abdominal Fat aspiration
- Biopsy
  - Abdominal Fat aspiration
  - Rectal biopsy
  - Gingival biopsy

kidney biopsy is done only when kidney involvement is suspected.

Stains

1. H & E → Pink homogenous appearance
2. PAS → ~~PA~~ PAS +ve · diastase resistant
3. CONGO RED → Most important stain *
   - Light Microscopy (LM) → Brilliant Pink Colour of amyloid.
   - Polarized light → Apple green birefringence → is due to β pleated sheet conformation of amyloid.

Metachromatic stains

Crystal violet & methyl violet.
↓
Megenta coloured amyloid.

Thioflavin T AS
→ UV light → Secondary fluoresence.

Immunohistochemistry

ORGAN — Site

Liver ⟶ 1st site is Space of Disse & causes pressure atrophy of hepatocytes.

ITO
↓
Storehouse of Vit A & synthesis collagen.

Kidney ⟶ 1st site is Mesengial matrix

Walls of capillaries of glomerulus, arteries ,, peripherial capillaries

Sub endothelial deposits.

Heart ⟶ Sub endo cardium

Between the Myocardial fibres

Will cause Arrythemias (bundle branch block)

Restrictive Cardio myopathy

C.C.F

GIT ⟶ Any where from mouth to anus.

Tongue — Macroglossia
— Amyloid tumor of tongue.

Spleen — Sago spleen
— Lardaceous spleen

SAGO SPLEEN Amyloid deposition in white pulp in lymphoid follicles in cortex.

deposits in Cortex / white pulp

⇓

Spleen has granular surface

On cross section
⇓
Gray Translucent bodies like grains of Sago.

LARDACEOUS SPLEEN

Amyloid deposition in walls of splenic sinusoids (& Red Pulp) ⟶ produces large map like areas of amyloidosis

LARD → animal Fat.

IMMUNITY

⇒ Self defence

Innate Immunity

- First line of defence
- Function - Prevention & erradication of infection.
- Innate immunity is non specific
- Innate immune response doesnot become better with each exposure
- No memary cells

Components

1) Epithelia - Skin, GIT, Resperatory Tract.
   - Mech Barrier
   - Produce antimicrobial substances eg: defensins
   - Intraepithelial lymphocytes

Adaptive Immunity

- 2nd line of defence
- Function - eradication of infection
- Adaptive immunity is specific
- Adaptive immunity response becomes better with each response
- Memory cells present.

Components

T lymphocytes
↓
Cell mediated immunity

B lymphocytes
↓
Humoral immunity

2) **Cells** -

- Neutrophils } Extra cellular bacteria
- Macrophages } & fungi

- NK cells – First line of defence against virally infected cells and tumor cells.

- Dendritic cells.

- Mast cells.

3) **Plasma proteins**

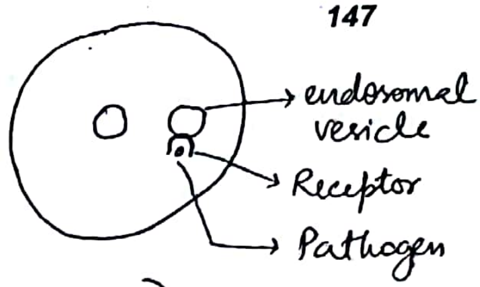
- Mannose binding lectin
- CRP
- Complement < Lectin pathway / Alternate pathway
- Lung surfactant

Pattern recognition receptors.

Cells that participate in innate immunity have receptors that recognize microbial components that are shared by microbes. These are called Pattern Recognition Receptors.

Location

Plasma membrane ✓
Endosomal vesicles ✓
Cytosol ✓



NF- kappa beta Pathway activated

↓

Cytokines secreated

↓

Recruitment of neutrophils & macrophages

Interferon regulatory factorss are produced (IRF)

↓

Stimulate production of antiviral cytokines e.g Type 1 interferons

↓

gets rid of viruses

Pattern ~~Rago~~ Recognition receptors.

① Toll like receptors - (TLRs)

Found on — Plasma membrane / Endosomal vesicles

11 TLRs are recognized Till date

e.g TLR-2 — gram +ve bacteria / Fungus / Leptospira

TLR-4 — gram -ve bacteria

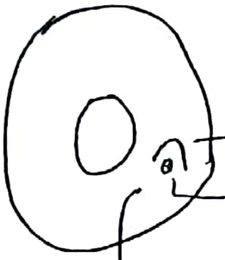
Founder member "TOLL" discovered in Drosophillia

2) NOD like receptors (NLR)

Location- cytosol

Recognize wide variety of substances

- Microbial substances e.g.; Shiglla & Salmonella
- Metabolic biproducts e.g.; uric acid
- Recog. ion disturbance
- Necrotic cells.



Inflamasome (Multi protein Complex) → activates Caspase I (IL-1β converting enzyme)

↓

IL - 1

↙ Fever ↘ Inflammation

⇒ Gain of function mutation in NLR gene leads fever periodic fever syndrome called <u>auto inflammatory syndrome</u> which responds to treatment with IL 1 antagonist.

③ C-type Lectin receptors

Found on plasma membrane

Detects Fungal glycans and leads to inflammation → gets rid of fungi.

④ Rig like receptors

Found in cytosol

Detects viral nucleic acid

↓

IRF are produced

↓

Type I interferons.

⑤ 7 transmembrane G protein coupled receptors

Recognize bacterial products $\bar{c}$ N formyl methionyl residues → stimulates chemotaxis.

⑥ Mannose Receptors

Recognize mannose sugar in bacterial wall

↓

Stimulate phagocytosis.

Adaptive Immunity.

T cells (CMI)

B cells. (HMI)

Lymphocytes are antigen specific

Mature lympocytes that have not encountered antigen or immunologically inexperienced are called

Naive Lymphocytes

Effector cells

Function - eliminate microbe

Memory cells

Live in a state of heightened awareness

Rapidly combat the microbe if it returns.

Lymphocytes are antigen specific - Lymphocytes of same specificity form a clone.

When an antigen enters, it selectively recruits antigen specific clone, this is called **clonal selection**.

Natural killer cells (NK cells)

Non B & Non T cells

Do not have TCR/BCR.

⇛ Also called large granular lymphocytes.

Function.

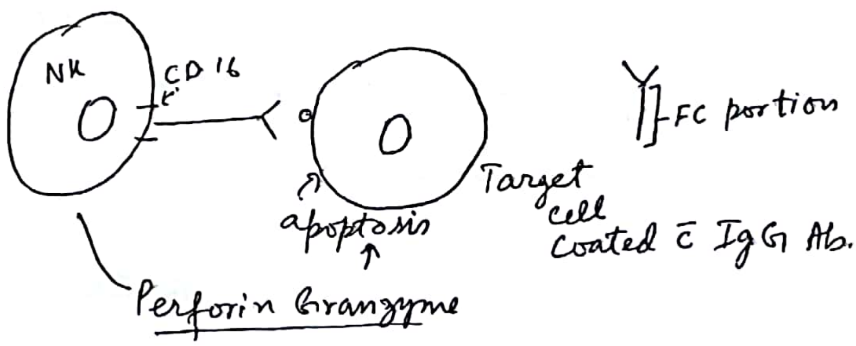
① Innate Immunity First line of defence against virally infected cells and tumor cells.

② Adaptive immunity Play a role in ADCC (antibody dependent cellular cytotoxicity).

Location → Constitute 5-10% of P. B Lympocytes.

Markers → $\boxed{CD\ 16}$ → FC receptor for IgG Ab.

$\boxed{CD\ 56}$ → Function Not known.



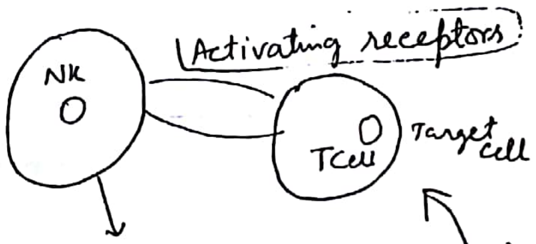
FC Portion of IgG Ab fits into CD16 on NK cells. NK cells release perforins & granzyme & Kill the Target cell.

Role in Innate Immunity

- kills virally infected cells & tumor cells.
- NK cells are not MHC restricted.
- NK cells have 2 types of receptors.

(1) Activating receptors
Activate NK cells to kill the target cells.

- Belong to NKG2D family.



Perforin granzyme killing

- NK cells attach to target cells by activating receptors & kill the target cells by Perforin granzyme dependent killing.

(2) Inhibitory receptors.
Prevents NK cells from killing Normal cells.

Inhibitory receptors belong to

CD 94
family of Lectins

KIRS (CD96)
(killer cells Ig like receptors)

Cytokines produced by NK cells – IFN gamma
↓
activates macrophages by classical pathway

Cytokines that regulate NK cell activity

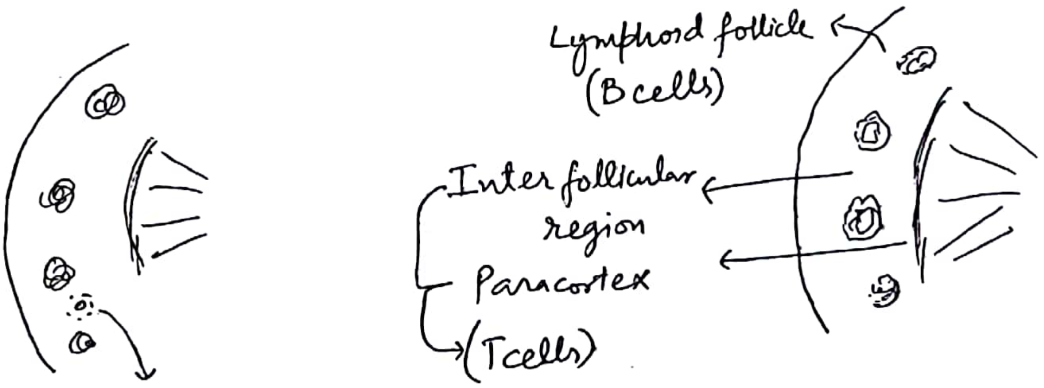
IL2, IL15 – Stimulates NK cell proliferation.

IL12 – Activates killing & secreation of IFN gamma by NK cells

T cells

Play a role in cell mediated immunity.

Location – Peripheral blood
60-70% of P.B. lymphocytes.



Periarteriolar sheaths of spleen

⇒ Found in Paracortical, Interfollicular region of LN

⇒ Found in Paracortical region & periarteriolar sheaths of spleen

T cell Markers

TCR (T cell Receptor)

CD1, CD2, CD3, CD4, CD5, CD7, CD8, CD28.

TCR

- Antigen specific
- 2 types
  - αβ TCR
    - Found on 95% T cells
    - MHC Restricted.
  - γδ TCR
    - Found on 5% T cells.
    - Not MHC Restricted.
    - Found in the epithelia like skin, GIT, urogenital etc → provide protection against microbes that try to enter through the epithelia.
    - Do not have CD4 & CD8 on their surface. (γδ)

CD3

⇒ Signal transduction is function

→ Lineage specific T cell marker

⇒ Pan T cell marker

CD7

Pan T cell marker

Not lineage specific.

CD4 }
CD8 } Found on two mutually exclusive subsets of Tcells.
(CD4 : CD8 :: 2:1)

CD1a Thymocytes & Langerhan cells.

Tcells

Helper Tcells
CD4 + Tcells
Master regulators

Function
- Help B cells to produce antibodies
- Help macrophages to kill or destroy microbes.

Cytotoxic Tcells
CD8 + Tcells
2nd line of defence against virally infected cells and
→ Tumor cells.
Kill Target cells by Perforin granzyme dependent killing.

Regulatory Tcells.
CD4 + Tcells.
FOX P3 +ve (transcription Factor)

Function.
Regulate immune response & prevent reaction to self antigens.

Helper T cells

- TH1
- TH2
- TH17

TH1

Cytokine produced – IFN gamma

[Biggest producer
Signature cytokine]

Function

IFN gamma
- Activates macrophages by classical pathway
- Stimulate B cells to produce IgG antibody.

Host defense against – Intracellular microbes

Role in disease – Chronic Auto immune disease
e.g: IBD, Psoriasis

TH2

Cytokines produced
- IL 4 (signature cytokine)
- IL 5
- IL 13

Function

IL4 stimulates B cells to produce IgE antibody
Activates Macrophages by Alternate pathway

IL5 – Stimulates B cells to produce Ig A Ab.
Activates mast cells & eosinophils.

IL 13 → Activates macrophages by alternate pathway
→ Activates epithelial cells to produce mucus

Host defense against – Helminthic parasites

Role in disease – Allergies.

TH 17

Cytokines produced

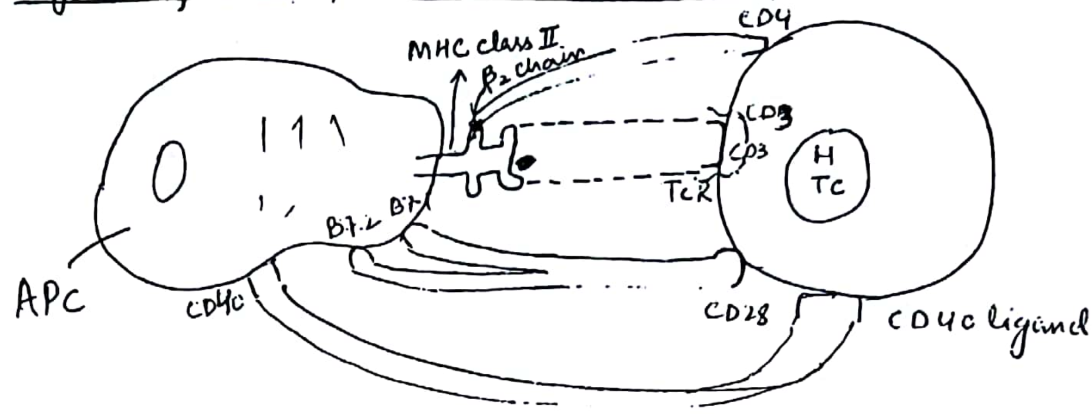
- IL 17
- IL 22
- Chemokines

Function – Recruitment of neutrophils and monocytes/macrophages

Host defense against – Extracellular bacteria & fungi.

Role in diseases – Chronic AID like IBD, Psoriasis & multiple sclerosis.

Signals for Helper T cell activation.



Signal 1

(a) TCR (Tcell) binds to antigen which is presented by APC in context of MHC class II

(b) CD4 of T cells attaches to β_2 chain of MHC class II

Signal 2 (also called- Co- stimulatory signal)

CD28 (Tcells) attaches to $\begin{bmatrix} B7.1\ (CD\ 80) \\ B7.2\ (CD86) \end{bmatrix}$ of APC

Signal 3

CD40 ligand (T cells) attaches to CD40 on APC.

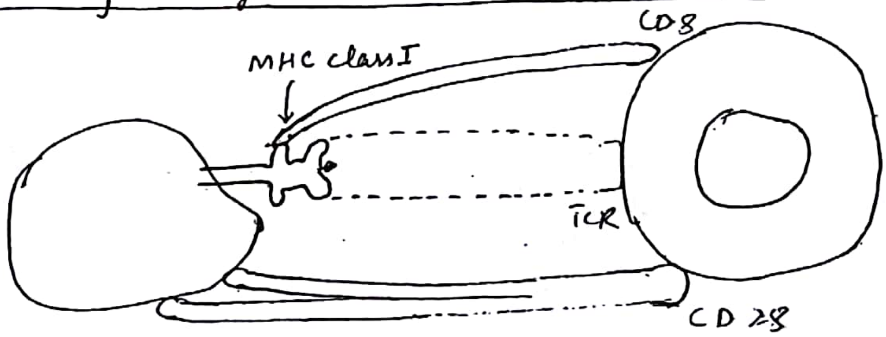
To stop T cell (helper) ~~cell~~ activation

by co inhibitory receptors < $\boxed{\text{CTLA-4}}$ / $\boxed{\text{PD-1}}$

Stops helper T cell activation

Belongs to CD28 family, blocks signals from TCR and CD28 & terminates T cell response.

Signals for Cytotoxic T cell activation.



Signal 1 (a) TCR (T cells) attaches to antigen that is presented by APC in context of ~~[illegible]~~ Class I MHC

(b) CD8 (T cells) attaches to α_3 chain of MHC class I.

Signal 2 Co stimulatory signal

CD28 (T cells) attaches to B7.1 CD80 / B7.2 CD86 of APC

→ CD8 T cell kills Target cells by Perforin Granzyme killing.

B cells

Humoral immunity

Sites [PB → Constitute 15-20% of PB lymphocytes
Found in lymphoid follicles in LN, spleen, Peyer's patches, BM, tonsils, etc.

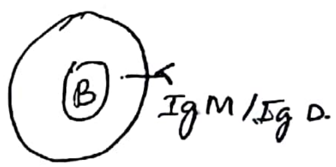
Bcell Markers → BCR (B cell Receptor)

CD19, CD20, CD21, CD22, CD23, CD10 (CALLA)

Igα (CD79a)   Igβ (CD79b)

BCR IgM/IgD antibody (Intramembranous)

- Antigen Specific.



Igα (CD79a)
Igβ (CD79b) } signal transduction (like CD3 of Tcells)

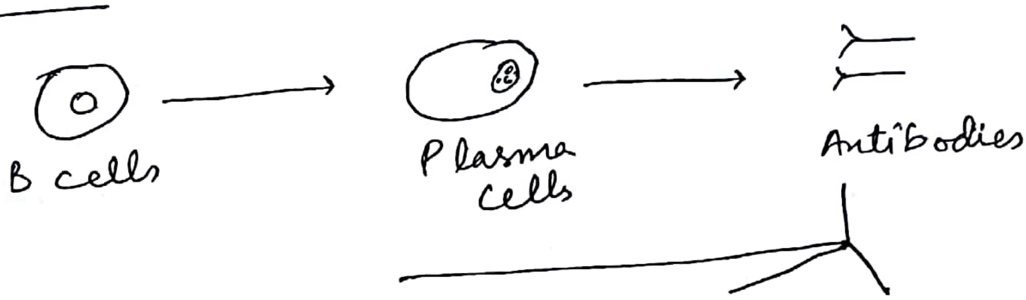
CD19 Pan B cell marker
Lineage specific

CD20 Lineage specific IHC

CD21 [Compliment Receptor (CR_2)
EBV receptor

CD40 site where B cells receive signals from Helper T cells.

Function



Neutralization

Antibody binds to the microbe & this microbe cannot infect any other cell.

Opsonization

IgG
Macrophage

Macrophages remove Opsonysed microbes

Complement fixation

Complement
↓
C5-9 (MAC)
↓
lysis of the bacteria.

↳ IgG antibodies cross placenta and provide passive immunity to new born.

↳ IgE antibodies → Parasitic infection

↳ IgA antibodies are produced in the mucosal surface → provide protection on mucosal surface.

2 pathways for Ab production by B cells.

T Independent Pathway

Polysaccharide & lipid antigen occupy a number of antigenic deterinants (ie BCR) on B cells

↓

B cells ⟶ PC → IgM

No help of T cells (HelperT) is taken.

T Dependent Pathway

Protein antigens stimulate B cells which bind to HelperT cells and then B cells produce

↓

IgG
IgA
IgE

Class of Ab

Help from Helper T cells is taken.

This is called iso type or class switching.

Helper T cells also stimulate B cells to produce antibodies with high affinities for antigen. This is called as affinity maturation.

Dendritic cells

- Antigen presenting cells – Best APC.

Best APC because

① Located at the Right place where antigens are encountered.

② They have fine hair like processes that trap antigens.

③ Each Rich in MHC class I & II.
& can present antigent to — CD4 T cell, CD8 T cell

④ They are also rich in co molecule B7.1 & B7.2

Dendritic cells → Interdigitating DC, Follicular DC

Interdigitating DC

Present Ags to T cells

Location –

Skin Langerhan cells.

Interstitia of organs e.g Lungs, GIT, heart liver. etc.

Langerhan cells

CD1a

S100

Langerin (CD207)

Follicular DC

- Present Ags to B cells
- Location – In centre of Lymphoid follicles

E/M Birbeck granules in their cytoplasm

- Zipper like
- Tennis racquet like.

MHC antigens

RBC do not have MHC Ag.

MHC Class I

HLA A

HLA B

HLA C

⇒ Found on all cells except RBCs

CD 8 T cells mount an immune response in context of class I MHC.

MHC class II

HLA DP

HLA DQ

HLA DR.

Found on APC i.e;

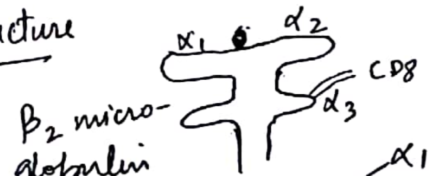
dendritic cells

B lymphocytes

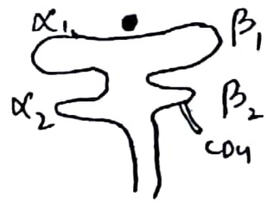
Monocytes/Macrophages

CD4 T cells mount an response in context of class II MHC.

Structure



β micro globulin + $\alpha \langle \alpha_1, \alpha_2, \alpha_3$



$\alpha \langle \alpha_1, \alpha_2$

$\beta \langle \beta_1, \beta_2$

Antigen binds in the cleft b/w α_1 & α_2

α_3 is the site of CD8 attachment.

Antigen binding cleft is b/w α_1 & β_1

β_2 is the site of CD4 attachment.

Genes for MHC antigen are found on the short arm of chromosome no. 6.

APC

Professional APC
- Dendritic cells
- Monocytes & Macrophages
- B cells

Non Professional APC
- Thyroid follicular cells
- Thymic epithelial cells
- Fibroblasts
- Glial cells
- Endothelial cells.

Hypersensitivity Reactions

Injurious immune reactions are called Hypersen-
-sitivity Reactions.

Type I

Rapidly occuring reaction which occurs within minutes of binding of antigen to IgE antibody on the mast cells in a previously sensitized individual.

eg:

→ Localized (ATOPY)
[Genetic predisposition]
Bronchial Asthma, Hay Fever
Allergic conjunctivitis, dermatitis
& allergic rhinitis
Food allergies

→ Systemic

Anaphylactic Shock

Drugs
e.g Penicillin

Hormones

Antisera

Bronchial Asthma

Pollen Antigen binds to APC (dendritic cells)

↓

APC presents Ag to Naive Tcells

↓

$\boxed{\text{TH2 Subset}}$

↓

TH_2 binds to B cells

↓

B cells ⟶ PC ⟶ IgE Ab

IgE antibodies attach to the Fc receptor on mast cells.

↓

Pollen Ag causes cross linking of IgE antibody on Mast cells

↓

Mast cells degranulate & release Mediators

↓ Preformed/Primary

Brings about initial phase of Bronchial Asthma

C/F →
- Vasodilation
- ↑ Vascular permeability
- Bronchospasm.

⇒ Histamine, Lysosomal enzymes (Proteases) serotonin.

↓ Newly synthesised or Secondary mediators

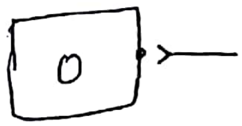
- Bronchospasm
- Leukocytic infiltration
- Epithelial damage

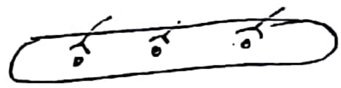
⇒ PAF, AA mediators (LTB4, LTC4, LD4, PGD2) Cytokine, Chemokine

Type II

- **Antibody Mediated** (IgG/IgM)
- Brought about by antibodies which are directed against fixed antigens

Antigen can be fixed on cell membrane
or
Connective tissue.





(a) - Antibody attaches to fixed antigen on cellmembrane & causes destruction of Target cell.

(Or)

(b) - Antibody attaches to fixed antigen & causes disregulation of function of target cell.

e.g Grave's Disease
Myasthenia gravis

Type V (in Micro)
& Type II here

Destruction by
- Opsonisation
- Compliment fixation

Antibody attaches to Basement membrane
↓
Activate Compliment
C3a C5A
↓
Enzymes released
↓
Breakdown of Basement membrane
↓
Good Pasture Syndrome

eg. AIHAnemia.
AI granulocytopenia
AI Erythroblastosis fetalis.
Mismatched blood Transfusion reaction.

Pemphigus vulgaris

Pernecious anemia (antibodies against Parietal cells)
↓
No IF
↓
Meg. Anemia.

Acute Rheumatic Heart disease

Type III
[Called Immune complex disease]

Antigen is not fixed.

3 Stages

(1) Formation of immune complexes (IC)

Antigen $\xrightarrow{\text{5days/1week}}$ IC

Most pathogenic complexes are
- Small/medium size
- Found in antigen excess.

(2) Settling down of IC

Favoured site
- Kidneys
- Joints
- Walls of Blood vessels
- Skin, etc

(3) Tissue destruction by Acts

- Activation of Complement
- Activation of Hageman factor.

→ Tissue destruction

eg.
- → Localized
  - → Arthus reaction
  - → APSGN, GN (T. pallidium)
  - → Reactive Arthritis (Yersinia)
- → Systemic
  - (1) → Serum sickness
  - (2) → SLE
  - (3) → PAN (Medium vessel vasculitis & IgG containing IC are deposited in vessel wall).
  - (4) → HSP (Small vessel vasulitis IgA containing IC are deposited in vessel wall)

SLE → All visceral lesions are type III rxn (IC disease)
→ All hematological lesions are Type II
- AIHA
- AI granulopenia
- AI Thrombocytopenia

Type IV

Brought about by T cells (Cytokine mediated)

CD4 T cells

Delayed HR

e.g: Tuberculin Reaction
Immune granulomas
Contact dermatitis

CD8 T cells

Destruction of Virally infected cells & tumor cells.

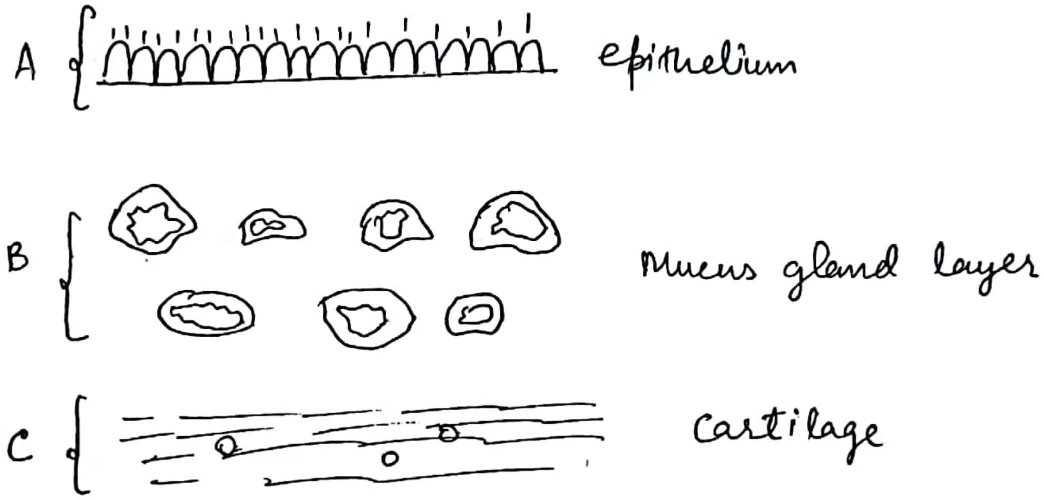
Both CD4 & CD8 ⇒

1. Rheumatoid Arthritis
2. IBD
3. Type I DM
4. Psoriasis
5. Multiple sclerosis.

Acute cellular graft Rejection

Graft versus Host disease.

Respiratory system



Reid's Index = $\frac{B}{A+B+C}$ [N = 0.4; ↑ sed = Mucus gland layer]

Asthma - Airway Remodelling

1. Sub Basement membrane fibrosis [Type III and Type I collagen]
2. Mucus gland hyperplasia
~~3. Goblet cell metaplasia~~
3. Goblet cell metaplasia
4. Smooth muscle hypertrophy + hyperplasia

ARDS

[Respiratory failure occuring ē in 1 wk of a known clinical insult ē B/L opacities on chest imaging, not fully explained by effusion, atelectasia, cardiac F. or F. overload]

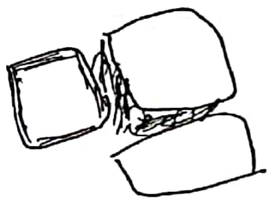
M/C/C — Sepsis > Pneumonia

IL-8, IL-1 and TNF α is involved.
↓
(Chemokine for neutrophils) → So, neutrophils are M/I culprit responsible for ARDS.

→ IL 8 rises ē in 30 minutes of injury

→ Cells damaged - endo & epithelial cells

leakage of fluid + cellular debris + leaked plasma proteins



↓

Hyaline
↓
Along the alveoli
↓
making membrane

→ Diffused alveolar damage (DAD) - Histological Hallmark

Coal worker's Pneumoconiosis

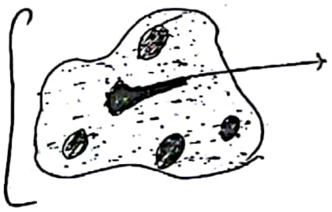
→ Pneumoconiosis → To describe non-neoplastic lung reaction to inhalational if mineral dust mainly at workplace ; also includes organic as well as inorganic particulates, chemical fumes & vapours

MC - Silicosis (quartz)
↓
Collagenous nodule in lung  → egg shell Calcification (lymph nodes classically)

Asbestos Related Disease

1. Pleural Plaque (MC manifestation)
2. Pleural effusion
3. Diffuse Pul. fibrosis → [Asbestosis]
4. Lung Ca – [MC Ca in asbestos exposure]
5. Mesothelioma → (Most specific Ca) (25-40 yrs)
6. Laryngeal Ca
7. Ovarian Ca
8. Colon Ca

(7, 8) → a Latent period ↓ (10-15 yrs)



ASBESTOS BODY [Coated c̄ Iron & Proteinaceous material
Iron ↓ Perls' Stain +ve

Any other particle coated c̄ Iron & proteinaceous material
↳ FERRUGINOUS BODY

In Anthraconosis → Pigment laiden macrophages are seen

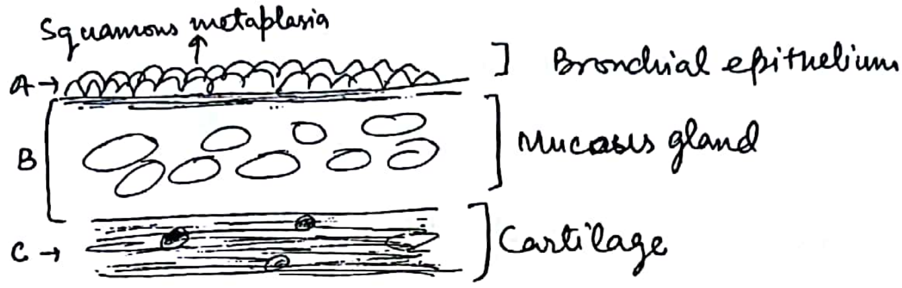
Coal Macule → Aggregated Macrophages (1-2mm)
↓
Coalese to form Nodule
↓
Progressive ~~massive~~ massive Fibrosis of lung parenchyma

↳ Risk of cancer is more in domestic use of coal i.e; Bituminous coal.

Chronic Bronchitis

MC cause - Smoking

Earliest change - MUCUS HYPERSECREATION



REID's Index $= \frac{B}{A+B+C}$

↓

done in autopsy (N) = 0.4

↑ed in → ↑ Mucous gland.

Manifestations in Pneumoconiosis

1. Localised fibrous plaques, or rarely diffuse pleural fibrosis (M/C menifestation)
2. Recurrent Pleural effusion
3. Parenchymal interstitial Lung fibrosis (Asbestosis)
4. Lung Ca (MC)
5. Mesothelioma (most specific)
6. Laryngeal, ovarian, colon Ca.
7. ↑ risk of autoimmune disease
8. CVS disease.

LUNG Ca

Histological classification (WHO 2015)

① Adeno Ca
- Acinar, papillary, Micropapillary, Solid, lipidic, predominant, Mucinous subtypes.

② Sq cell Ca
→ Non keratinising
→ Keratinising
→ Basaloid

③ Large cell Ca

④ Neuroendocrine Ca
- Small cell Ca
- Large cell neuroendocrine Ca
- Carcinoid Tumor

Adeno Ca

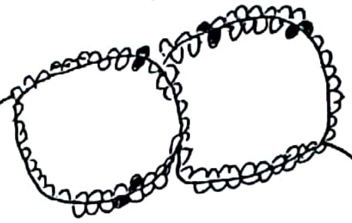
- Pre invasive lesions → Atypical Adenomatous Hyperplasia
  → Adeno Ca insitu
- Minimally invasive Adeno Ca
- Invasive Adeno Ca

Adeno Ca Insitu (AIS)

Previously known as Bronchoalveolar Ca.

→ < 3cm
NO stromal invasion
NO Pleural invasion
NO Lymphovascular invasion.

⇒ Butterfly sitting on Fence Appearance



(No desmoplastic Stroma)

LEPIDIC PATTERN OF GROWTH

Minimally Invasive

< 5mm stromal invasion & < 3mm in size.

No pleural invasion

No lymphovascular invasion.

→ Basophilic Psamoma bodies

Eosinophilic

Squamous cell Ca
- → Keratinising (New)
- → Non keratinising
- → Basaloid (New)

Neuroendocrine Origin

- Histology
- IHC
- E.Microscopy.

Histology

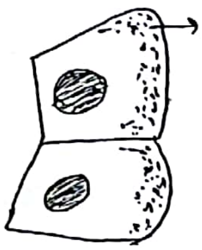
Salt & Pepper Chromatin/ Stippled Chromatin

In High Gr- Hyperchromatic

IHC

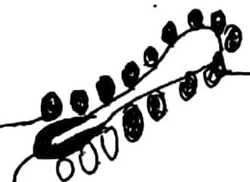
Synotophysin
Chromogranin
& CD56
CD57
] +ve

E.Microscopy



Small cell Carcinoma

[Oat cell carcinoma]



Salt pepper appearance

Azzopardi effect
(DNA encrustation on vessel wall)

Small cells
↑ N:C ratio
Round to oval nuclei
Stippled chromatin

Tumor cells have a high turnover & and are fragile ⟶ disrupt easily
↓
Their DNA gets encrusted upon vessel wall [BLUE]

Purplish Blue Powdery discolouration of vessel walls
⇓
Azzopardi effect

Carcinoid [Carcinoma like epitheloid tumor]

	Grade	Mitotic count
Typical	I	< 2/10 HPF
Atypical	II	2-10/10 HPF

HPF = High Power Field
~ 40X

Abundant cytoplasm (Pink cells)

Monotonous round nuclei with stippling

Fibrous Septae making nests of ~~cytoplasm~~ tumor cells.

Tumor genetics

Small cell	Squamous cell	Adeno
Rb mutation	P53 mutation	EGFR mutation > amplification
P53 mutation	EGFR ~~mutation~~ amplification (more than mutation)	KRAS mutation
myc amplification	FGFR2 amplification	ALK rearrangement

Amplification is checked by FISH
↓
Overexpression is checked by IHC

Genetic change

Mutation → Change in DNA sequence (PCR sequencing) → No Protein / Abnormal protein (IHC)

Rearrangement → Change in locality of gene → Translocation / Inversion → (karyotyping / FISH)

Due to Translocation & inversion

① gene 1 + gene 2

↓ Transcription

(Fusion Transcript)

(Fused mRNA) (RT PCR)

↓

(ONCOPROTEIN)

(IHC) [↑ed function than before] → e.g BCR ABL

or

[↓ed function than before]

② gene 1 gene 2 (promoter)

↓

gene 1 ↑↑↑ed (FISH)

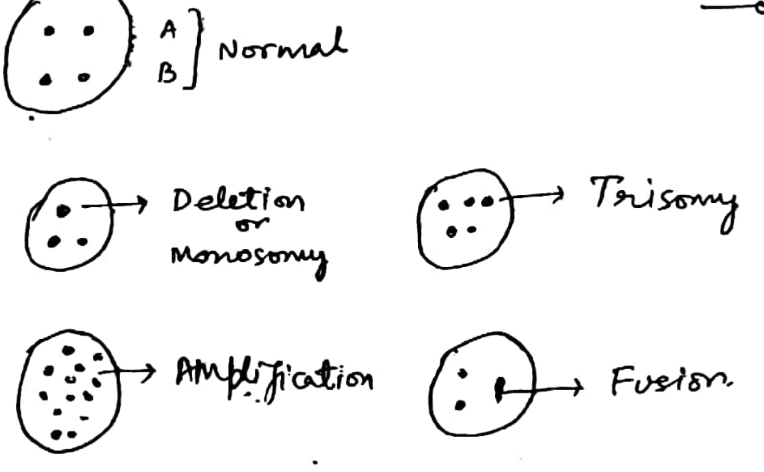
(overexpressed)

↓

Overexpression (IHC)

FISH

FISH INTERPRETATION



ALK

[Anaplastic lymphoma Kinase]
Receptor Tyrosine kinase

- → Rearranged →
  - → Anaplastic large cell lymphoma (ALCL)
  - → ~~Lymphoma~~ Lung Adeno Carcinoma
  - → Inflammatory Myofibroblastic tumor
- → Mutated
  - Neuroblastoma
  - Hereditary Haemorrhagic Telangiectasia Type II

In lung Adenocarcinoma

EGRF & ALK can be targetted
[EGRF & KRAS are mutually exclusive]
In non smokers & women → EGRF mutation.

IHC of lung Cancer

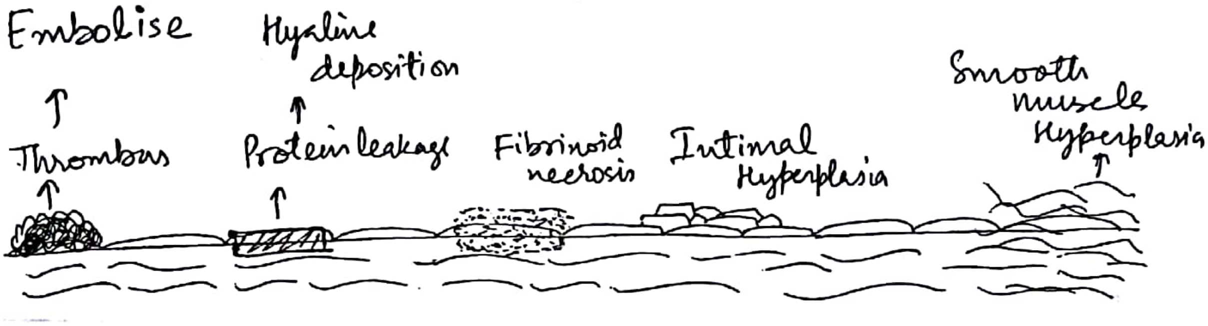
	Small cell	Squamous	Adeno
Thyroid Transcription Factor (TTF-1)	+	−	+
NAPSIN-A	−	−	+
Other markers	Synaptophysin Chromogranin CD56/CD57	p63	CK7 + CK 20 −

	Mesothelioma	Adenocarcinoma
PAS	–	+
Carcinoembryogenic Antigen	–	+
TTF1	—	+
NAPSIN	—	+
CK7	+	+
CK5/6a	+	—
Calretinin	+	—
WT1	+	—
Electron microscopy	Long slender microvilli	Short Stubby villi

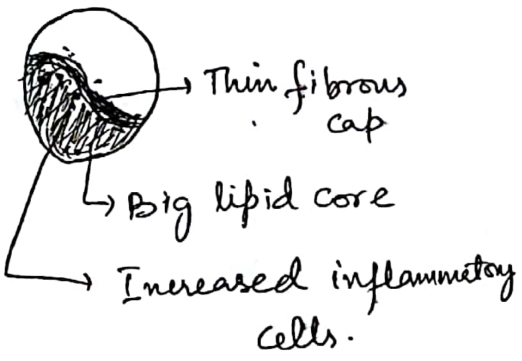
Adenocarcinoma	Squamous cell Carcinoma	Small cell Carcinoma
- Most common lung Cancer world wide In women In non smokers	- Most common lung Cancer in India in Smokers	- Strongest association with smoking.
- Peripheral Location	- Central location	- Central location
- Most common gene mutated is KRAS	- Most common para-neoplastic syndrome is - Hypercalcemia	⇒ Most common Para-neoplastic syndrome is SIADH.
⇒ Most common Para-neoplastic syndrome is Hematological	⇒ Cavitation (also in large cell ca)	⇒ SVC obstruction
⇒ Clubbing is seen	⇒ Pan coast tumor ↓ HORNER's Syndrome	LAMBERT EATON Synd.
		⇒ Worst Prognosis
		⇒ Max Risk of Metastasis
		⇒ Clubbing is Rare.

Lung cancer most commonly metastasises to BRAIN & most specifically to Adrenals.

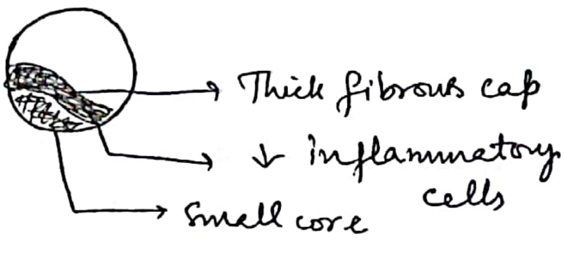
VASCULAR PATHOLOGY



① Atherosclerosis : Atheroma/ Atheromatous plaque.



VULNERABLE PLAQUE



STABLE PLAQUE

② Monckeberg Medial calcific Sclerosis

- No luminal obstruction
- clinically insignificant

Q 3. Arteriosclerosis :→ Hypertensive changes.

Site → arterioles

Benign

↓

Hyaline Arteriosclerosis

- Wall thickening
- Narrowing of lumen
- Kidney → Benign Nephrosclerosis
- Granular kidney : Leather Grain Appearance

~~Kidney e Malign~~

Malignant

↓

Hyperplastic Arteriosclerosis

PAS stain  → Onion skin appearance

↓

New Basement membrane

Kidney → Malignant Nephrosclerosis

Pinpoint Hemorrhages

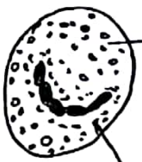
↓

Flea bitten Appearance

Vasculitis → Infectious
Vasculitis → Non Infectious (Immune)

Mechanisms

1. Anti endothelial or Anti Smooth muscle antibody
2. Immune complex deposition
3. T-cell Mediated Response (granulomatous)
4. ANCA (Anti neutrophilic cytoplasmic Antibody)



→ cytoplasmic : Proteinase-3 (PR 3)
(diffused)
Antibody against these (c ANCA or PR3 ANCA)

→ Perinuclear - Myeloperoxidase (MPO)
Antibody against these (p ANCA or MPO ANCA)

Leucocyte Activation → surface exposed to these granules
↓
If ANCA Present (Susceptible individuals)
↓
Binds to the surface granules → Degranulation of Neutrophils
↓
Enzyme Leakage → Tissue damage.

CHAPEL HILL CONSENSUS CLASSIFICATION

Large Vessel
- Gaint cell arteritis (Pulse less disease)
- Takayasu arteritis

Medium vessel
- Polyarteritis Nodosa
- Kawasaki disease

Small vessel

Immune complex vasculitis
- IgA vasculitis (Henoch schonlein Purpura)
- SLE

ANCA associated vasculitis
→ Granulometosis with Polyangitis (wegner's Granulomatosis)
⇒ Eosinophilic Granulometosis with Polyangitis (Churg Straus Syndrome)
⇒ Microscopic Polyangitis

& Variable vessel
→ Becket's disease
→ Cogan syndrome.

Giant cell Arteritis	Takayasu Arteritis
> 50yrs	< 50yrs
Head & Neck arteries Temporal – Headache (MC) Facial → Jaw claudication Ophthalmic → Blindness [Most specific]	**Aorta & Branches** ↳[Aortic arch] Most commonly : Subclavian ② Common carotid ③ Abdominal aorta ④ Renal ⑤ Aortic arch or root ⑥ Coeliac axis ⑦ Coronary ↳(least common)
Diagnosis Biopsy – Segmental involvement ↓ 3 - 5cm → adequate biopsy Required. or at least 1 cm	**Diagnosis** CT Angio Arteriography.

Etiology

Anti endothelial & Smooth cell antibodies (?) — 〃

Cytokine mediated Response — 〃

Granulomatous Inflammation — 〃

Histopathology

[van Geison Stain for Elastin]

Polyarteritis Nodosa (PAN) [MC - Kidney involved]

- P AN
- P ATCHY → (entire circumference of vessel wall is not involved)
- P ULMONARY ARTERY SPARED [Bronchial vessels may be involved]

Features

Transmural (Through out wall) : Aneurysms, Fibrosis

Necrotising : Fibrinoid Necrosis

Inflammation : < Acute / Chronic] All stages of activity can be seen at the same time (ongoing Injury)

Associated c̄ Hep. B. → Immune complex deposition.

⇒ The pathology in the kidney in classic polyarteritis nodosa is that of arteritis without glomerulonephritis

⇒ May involve bronchial vessels but not pulmonary vessels.

⇒ Renal & visceral arteries are very commonly involved. (+ Muskuloskeletal)

⇒ No granulomas. No eosinophilia

Graint cell Aortitis → Aortoartitis

Kawasaki Disease

- < 5 years
- Anti endothelial cell antibodies ⊕
- Acute Necrotising Vasculitis (Transmural)
- Febrile illness
- Strawberry Tongue
- Cervical lymphadenopathy
- Mucocutaneous ulcers

⇒ MUCOCUTANEOUS LYMPH NODE SYNDROME.

Strawberry cervix ↓ Trichomonas infection

Cholesterolosis → Strawberry Gall bladder

- Most common artery involved ⇒ **Coronary** ↓ Thrombosis, M I, Aneurysms → MC cause of cardiac mortality in Children.

ANCA associated

Wegener's	Churg Strauss	MPA (Microscopic Poly-Angitis)
- c ANCA > p ANCA	- p - ANCA	- Hypersensitivity or p ANCA
- URT + LRT involvement	- Asthma	- LEUCOCYTOCLASTIC VASULITIS ↓ (WBC - Breakdown) ↳ Apoptotic Neutrophils.
- Renal involvent	- Allergic Rhinitis	- No granulomas
- Necorotising &/or granulomatous vasculitis	- Nasal polyps	- Fibrinoid Necrosis
	- Peripheral eosinophilia	
	- Necrotising &/or granulomatous vasculitis.	

URT - Upper Respiratory Tract involvement

Vascular Tumors

↓

Endothelial cell origin   IHC → PECAM / CD31

Benign

Hemangioma

Capillary Hemangioma

Cavernous Hemangioma

Boderline

Kaposisarcoma

$\boxed{\text{HHV-8}}$

1. Classical
2. Endemic (Africa)
3. Transplant associated
4. Immunodeficiency associated

Malignant

Angiosarcoma

1. Hepatic

   Exposure to Poly vinyl chloride & Thorium contrast dyes

2. Cardiac

   Most common primary cardiac malignancy in adults.

Microabscesses (Misc)

- Neutrophil Microabscesses in TAO
- Neutrophil crypt abscess in IBD
- Pautrier's Microabscess (Tumor lymphocytes in mycosis Fungoides)
- Munroe Microabscess of Neutrophils in Psoriasis

Thyroid Cancers

Follicular origin

Papillary : Most common

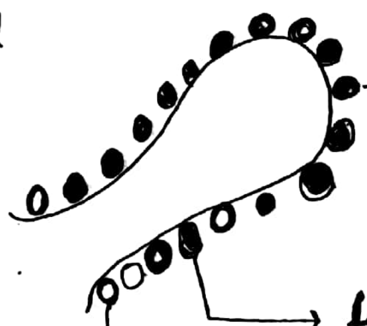
|

Parafollicular origin
(c-cells)
↓
CALCITONIN
↓
Medullary
Amyloid (A cal)
CEA
MEN2 syndrome

Papillary Ca Thyroid

Most common subtype | overall
Children
Thyroglossal cyst
Post Radiation
Hashimoto Thyroiditis

⇒ Lymphatic Spread
⇒ Best prognosis



→ Nuclear Pseudoinclusions (artefact)

→ Longitudinal grooves
↕
Coffee Bean Nuclei
[Granulosa cell Tumor]
also found in
Brener Tumor
Chondroblastoma
Langerhan cell Histiocytosis

- Chromatin margination
- Optical clearing of nucleus

ORPHAN ANNIE - EYE NUCLEI

Follicular Ca

Most common subtype to arise in multinodular goiter

⇒ Capsular or vascular invasion

⇒ Cytology (FNAC) will not help

⇒ Histopathological examination (No biopsy is done) → Hge

↳ also in Testicular mass.

↓

[Hemithyroidectomy to be done]

⇒ Hematogenous spread (Bones & lungs)

Anaplastic Ca

Bizarre cells

Fibrosis ↑↑

Least common subtype

Most aggressive (rapid onset)

Worst prognosis.

Tumor genetics

POG → Proto onco gene

TSG → Tumor supressor gene

Rearrangement ← RET (POG) → Mutation → Medullary

Rearrangement ↓ Pappillary

↓

KRAS (POG) → Follicular & anaplastic Ca

KRAS → BRAF (POG), PI3K (POG)

V600E Point mutation (valine --> Glutamate) ← BRAF (POG)

PI3K (POG) → Follicular & anaplastic Ca

PI3K ⟷ PTEN (TSG) → Follicular & anaplastic Ca

↓

PAX-8 (POG)

↓

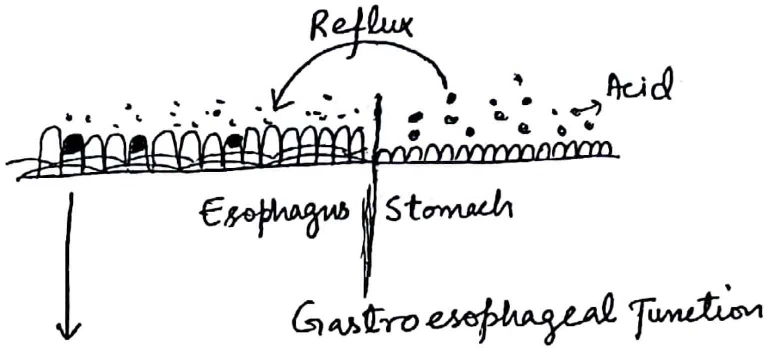
Follicular Ca (Rearrangement)

Q. Most common genes

Papillary : BRAF
Follicular : KRAS
Anaplastic : P53
Medullary : RET

GIT

Esophagus



↓

Columnar cells
+
Goblet cells → Acid mucin] → Alcian blue Stain (Q)

↓

Intestinal metaplasia → BARRETS Esophagus (Q)

Gross → Red velvety patches in esophagus.

Metaplasia → Displasia → Anaplasia (Irreversible)

↓

[Adeno-Carcinoma]

[Some serotypes of H pylori are associated c̄ decreased risk of esophageal adeno.ca. because they cause gastric atrophy → ↓ acid → ↓ Barret's esophagus]

Gastro Intestinal Stromal Tumor (GIST)

Origin Interstitial cells of cajal (Pacemaker cells in Muscularis propria)

Gross Nodular encapsulated tumor
Arising from the wall
Mass effects
Cut section → Grey Tan, Fleshy tumor with hemorrhage & necrosis

Strong ↓ Vimentin - Mesenchymal marker

Microscopy →
- Spindle cell (most common)
- Epithelioid
- Mixed

<u>Genetics</u>

<u>85%</u> $\boxed{\text{c kit}}$ mutation → AML, Seminoma, Mastocytosis

↓

$\boxed{\text{Imatinib}}$ → Inhibitor → Receptor Tyrosine kinase (RTK)

~ <u>8%</u> $\boxed{\text{PDGFRA}}$ mutations

[c kit & PDGFRA mutations are mutually exclusive]

In a small proportion of <u>non ckit</u> & <u>non-PDGFRA</u> <u>mutated</u> <u>GIST</u> ⟶ $\boxed{\text{SDH mutation}}$

↓

SDH Deficient (Succinate Dehydrogenase)

<u>Exclusively seen in</u>
- ↳ Gastric In Location
- ↳ Indolent Course (slow growing)
- ↳ Younger Population
- ↳ Imatinib Resistant
- ↳ <u>Part of Carney Stratakis</u> <u>Syndrome</u> [Paragangliomas] → Myxomas, Skin Lesions, Multiple endocrine Involvement

Carnary Triad → GIST, Pulmonary Condroma (Hamartoma), Paraganglioma.

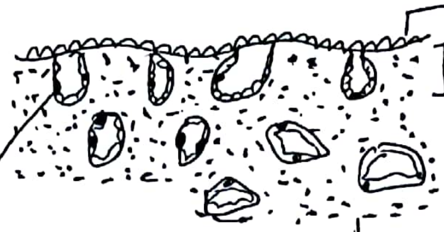
<u>IHC</u> → CD117 (ckit): +ve (Most sensitive)

DOG 1 : +ve (most specific)

<u>Prognosis</u> → <u>Location</u> (stomach: better)
→ <u>Size</u> (< 5 cm: better)
→ Mitotic count (Counted per 50 HPF)

Different cut offs for different sizes at different locations.

Celiac Disease



(CD8+) ®

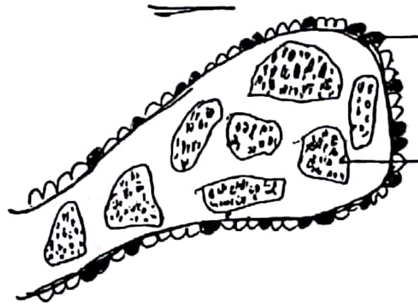
Risk of enteropathy associated T cell lymphoma is high (EATL) ®

- Adenocarcinoma Risk also increased because of crypt Hyperplasia
- Associated c̄ HLA DQ2 > DQ8
  - IgA Nephropathy
  - Dermatitis Herpetiformis

Whipple's Disease (Malabsorptive syndrome)

H&E

PAS



Intestinal polyps

Non Neoplastic

- Hyperplastic
- Inflammatory (MC)
- Hamartometous → Indigenous to that organ
  - ① Disorganised mass
  - ↳ now considered Neoplastic
  - ② Choristoma ↓ Normal tissue, Abnormal site

Peutz Jegher Polyp | Juvenile Polyp.

Neoplastic polyps

Adenomatous/Adenomas

DYSPLASIA

Peutz Jegher Polyp

Christmas Tree appearance

Multiple PJPs + Mucocutaneous melanosis

⇓

Peutz Jegher Polyp (STK 11 gene)

aka - LKB1

Juvenile Polyp

Solitary → No risk of cancer

Multiple → Juvenile polyposis Syndrome

↓ Slight risk of Cancer

BMPR 1A – (Primary Pulmonary Hypertension)

SMAD 4 – (Pancreatic Cancer)

Lymphoma

MC primary → MALToma (extranodal Marginal Zone Ly

↳ H Pylori associated

CD5 – , CD10 –

CD23 – , CD43 +

Adenomas/Adenomatous polyps

[Tubular](mc)

(Villous)

(Tubulovillous)

Famillial Adenomatous Polyposis (FAP)

APC gene mutation

Most commonly: Tubular Adenomas

Diagnosis > 100 polyps

Attenuated FAP < 100 polyps. (variant)

FAP + Skull Osteomas + Epidermoid cysts + Desmoid Tumor (Fibromatosis)	GARDNER SYNDROME

FAP + Meduloblastoma (More common) & Glioblastoma] Turcot syndrome

Colo Rectal Ca

Adenoma - Carcinoma Sequence.

Normal colonic epithelium $\xrightarrow[\text{APC}]{\text{loss of}}$ Hyper proliferation

(gate keeper of Colonic neoplasia)

Hyper proliferation $\xrightarrow{\text{epigenetic changes}}$ Early adenoma

Early adenoma $\xrightarrow{\text{KRAS \& NRAS mutations}}$ Late adenomas

Late adenomas $\xrightarrow{\text{Loss of P53.}}$ Adenocarcinoma

Others: EGFR, BRAF

Right sided colon	Left sided colon
Diameter is more → space for tumor to grow → Cauliflower/fungating/Polyploid	Diameter is less → No space to grow → Constriction (Napkin Ring constriction)
⇓ Loose consistency stools → easily pass through ↓ Causes erosion of tumor → Bleeding (Fecal occult blood test) ↓ Iron deficiency Anemia	Bulky stool → Cannot pass easily ↓ features of Intestinal obstruction

Hereditary Non Polyposis colon Cancer (HNPCC) or LYNCH SYNDROME

Defect in DNA Mismatch Repair gene (MSH2, MLH1)

↓

DNA mistakes accumulate → genomic Instability

MSI ← [Especially microsattelites] (Tandem sequences)

[Micro Satellite Instability]

Colon Cancers	Extracolonic Cancers
- Mean age ~ 40yrs	Endometrial (MC)
- Proximal to splenic flexure	Gastric
- Mucinous/signet ring cell morphology	Ovarian
- ↑sed lymphocytes in tumor	Transitional
- Better prognosis	Small intestinal

Pancreatic Carcinoma MC Subtype → Ductal Adenocarcinoma

Very aggressive

Procoagulant Tumor mucin + cellular debris

↳ DIC risk is increased

→ Non bacterial Thrombotic endocarditis

→ Migratory thrombophlebitis
(TROSSEAU SYNDROME) → also seen in Breast Ca, Gastric Ca, Lung Ca

Normal Pancreatic epithelium

KRAS ~90% (Poc)
CDKN2A/P16 ~95% (TSG) → (Protein)

Pancreatic epithelial Neoplasia (PanIN)

SMAD4, BRCA2, P53 — TSG

Adenocarcinoma

Most common gene Mutated - CDKN2A

Most common Protooncogene mutated - KRAS.

Most common TSG mutated → CDKN2A/P16

[KRAS changes occur earlier in the Progression]

Chromosome locations (Misc)

BRCA1 - 17q

P53 - 17p

APC - 5q

HFE - 6p

SERPINA1 - 14q

13q - BRCA2, Rb, ATP7B.

MET (Hepatocyte Growth Factor Receptor) - 7 chr. ↦ Papillary RCC

Mallory Dank Bodies

Mallory Dank Bodies/ Mallory Bodies

↓

Remnants of intermediate filaments. Q
following hepatocyte damage.

↓

cytokeratin Q

↓

CK 8/18 Q

→ Eosinophilic Inclusions.

Prognostic Markers for tumors

Stage	Lymph Nodes	Others
Esophagus	Breast (Axillary)	Metastatic Breast – ER/PR status
Stomach	Colon	Renal – Pathological Stage
Gall bladder	Penile (Inguinal)	Wilm's – Histology (Anaplasia)
Pancreatic	Head & neck	Soft tissue Sarcoma – Grade
Prostate		Melanoma – Depth
Testis		

Esophagus, Stomach, Gall bladder → T. Stage ↓ Depth of invasion.

$\boxed{\text{Barret's Oesophagus} \rightarrow \text{AdenoCa}}$

Early genetic Changes

- P53 (TSG)
- CDKN2A/ p16 (TSG)

Late genetic changes

- EGFR (erbb 1)
- Her 2 Neu (erbb 1)
- MET
- cyclin D_1

} Proto onco genes [POG]

Cardiac Pathology

Myocardial infarction

Coronary Artery Spasm → Hypoxia damage → Myocardial response (Ishemic Injury) (↓ ATP)

- ≈ 30 minutes Reversible (20-40min)
- Beyond - Irreversible

Infarction

TTC (Triphenyl Tetrazocium chloride)

- → +LDH → BRICK RED : Normal Viable myocardium
- → -LDH → PALE YELLOW : Recent infarct

Features	Onset
Onset of ATP Depletion	Seconds
Loss of Contractility	< 2mins
ATP Reduced to 50% of Normal[a]	10 min
to 10% of Normal	40min
Irreversible injury[a]	20-40 min
Microvascular injury	> 1 hr.

Valvular H.D.

	Morphologically	Special points
Mitralvalue Prolapse	Gross Balloning of value a/w Myxomatous degeneration	Marfan's Syndrome Mid systolic click
Rheumatic H.D	Verrucous vegetations along lines of closure [MAC cullum Plaques]	Pathognomic Aschoff nodule
IE (Infective Endocarditis)	Friable vegetations along lines of closure, invading Chordae Tendinae	Risk of Septic embolisation
NBTE (Non Bacterial Thrombotic endocarditis).	Large Fibrin clots along lines of closure ; No Invasion	Seen in Cancer Malnourished (Marantic Endocarditis)
LSE (Libmann Sack Endocarditis)	Vegetations on both sides of Cusp	SLE

Rheumatic Heart Disease

Q Molecular Mimicry

Acute Pancarditis < Myo, Endo, Peri

↓

Pathognomic feature is Aschoff Nodule which [can be seen in myo, endo, pericardium] Q

→ Plasma cells

→ Fibrinoid Necrosis

→ ~~Plasma cells~~ Aschoff gaint cells

→ Lymphoid cells

Modified Cardiac Histiocytes

↓

Anitschkow cell — wavy chromatin : Caterpiller cells

owl's eye appearance on cross section

Endocarditis

Verrucous vegetations along the lines of closure
↓
~~Body~~ warty

↓

Fibrinoid Necrosis
Inflammetory cells, Immune complex

↓

Regurgitation ← Damage the values

↓ Regurgitation Jets into Left Atrium

Overtime can cause Plaque formation especially in posterior wall of (L) atrium (when mitral value involved)

↓

Mac Callum Plaques

Chronic RHD

Due to inflammation
↳ Stenosis of valves
Due to fibrosis of cusps & Narrowing of orifice

Fish mouth / Button hole appearance

↓

Most commonly : $\boxed{\text{Mitral > Aortic}}$

Finding in eye → Superiotemporal dislocation of lens
⇓
MARFAN'S Synd

⇒ Owl's eye → CMV inclusions
→ R.H.D
→ Lymphoma

Fish mouth appearance


Floppy Mitral value
⇓
MVP

CNS & PNS Pathology

Schwanoma – Benign Peripheral nerve sheath tumor

↓

Most commonly – Cerebellopontine Angle Q

NF_2 associated

Variably cellular tumor

- Hypercellular compact areas → Antoni-A
- Hypocellular Loose areas → Antoni-B

→ Tumor nuclei are arranged in parallel to each other ⇒ TRUE PALISADING Q

→ Q VERROCAY BODY

CNS TUMORS

1 Glioma (M.C 1° tumor)
- Astrocytoma (MC) glioma
- Oligodendroglioma
- Ependymoma

2. Meningiomas (2nd m.c 1° tumor)

3. Embryonal Tumors
- Meduloblastoma (M.C 1° CNS malignancy in children)
- Atypical Teratoid/Rhabdoid Tumor (ATRT)

PNET is not included under this anymore WHO 2016

4. Neuronal Tumors (neurofilament +ve)

5. Lymphomas

6. Secondary Tumors (M.C CNS Tumors)
↳ (1° is Lung Ca MC)

ASTROCYTOMAS

4 grades | WHO 2016

4 grades	WHO 2016
I Pilocytic Astrocytoma	→ SEPARATE
II Diffuse Astrocytoma, III Anaplastic Astrocytoma	+ Oligodendroglioma → DIFFUSE GLIOMA
IV Glioblastoma	→ SEPARATE

Diffused glioma
↓
Histology → Grade II, III, oligodendroglioma

Histology → grade I, IV
Astrocytoma
↓
Glioblastoma

IDH ← mutations.

Pilocytic Astrocytoma

Most common 1° CNS Tumor in Children

Seen in Cerebellum

BRAF < Mutated (V600E) / Rearranged

Gross

↓

(Hemangioblastoma)

Micro

crystalline

& ROSENTHAL FIBRES

↳ Made of GFAP Proteins & other Proteins

(Non neoplastic reactive change)

Glioblastoma

Most common 1° CNS malignant in Adults

Seen in Frontal & Parietal Lobes

EGFR

P53

CDKN2A / p16

PDGFRA

Gross

- Tumor infiltrating normal brain

↓

crosses midline (Butterfly glioma)

- Areas of Hge & necrosis

Micro

Leaky vessels → Contrast enhancing

↳ Areas of necrosis (Pink) bordered by viable Tumor cells (blue dots)

↓

PSEUDOPALISADING NECROSIS

OLIGODENDROGLIOMA

Monotonus uniform tumor cells with Perinuclear Halo
[Fried egg appearance
(also in - Hairy cell Leukemia
- Mycoplasma)

Delicate anastamosing vasculature
[chicken wire vasculature]$^{\Omega}$

Chicken wire
↳ Fibrosis – Alcoholic liver disease
↳ Calcification – Chondroblastoma

Microcalcification : Imaging
↳ (Basophilic on H&E)

Genetics
- IDH1 IDH2 mutation
- 1p/19q codeleted
→ Favourable Prognosis

MEDULLOBLASTOMA [grade IV]

Mc site – cerebellum (Posterior fossa)
CSF dissemination → Drop metastasis

Small round blue cell tumor

→ Neurophil (cytoplasmic feature)
HOMER WRIGHT ROSETTE
↳ (PseudoRosette

(True Rossete has Nucleus)
↳ Ependymal Rosette

MENINGIOMA (I - III)

Arises from Arachinoid cap cells / Meningothelial cells

Dura based tumor (DURAL TAIL)

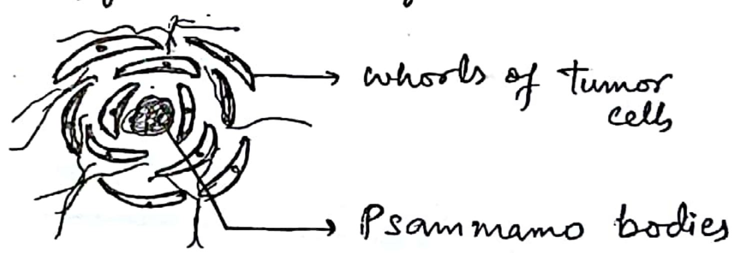
Easily Detachable

→ Causes reactive hyperostotic change in overlying bone.
↓
X Ray

Express PR ⇒ ↑ses in size during pregnancy.

Associated c̄ NF_2

Most common Histology is – Meningothelial



WHO 2016 → genetic incorporation into histology

Glioblastoma → IDH mutations (Isocitrate Dehydrogenase)

- ↓ Present
  - ↓ Glioblastoma, IDH mutant (better prognosis)
- ↓ Absent
  - ↓ wild type glioblastoma

Diffused gliomas

↓

IDH Mutation Status

- ↓ IDH Mutant
  - ↓ Depending on histology
    - looks like Astro → ATRX, P53 (IHC)
      - ↓ Diffuse Astrocytoma IDH Mutant
      - Intermediate Prognosis
    - looks like oligo → 1p/19q codeletion (FISH)
      - ↓ Oligodendroglioma 1p/19q codeletion & IDH mutant
      - Excellent Prognosis
- ↓ IDH wild
  - ↓ depending on histology
    - ↓ Diffused Astrocytoma, NOS
    - Oligodendroglioma, NOS
    - (Poor prognosis)

Schwanoma

Neoplastic Schwanomma cell

Cerebellopontine angle = Most Commonly

S100 +ve

NF2 associated.

Variable cellular areas

Antoni A

Antoni B

Neurofibroma

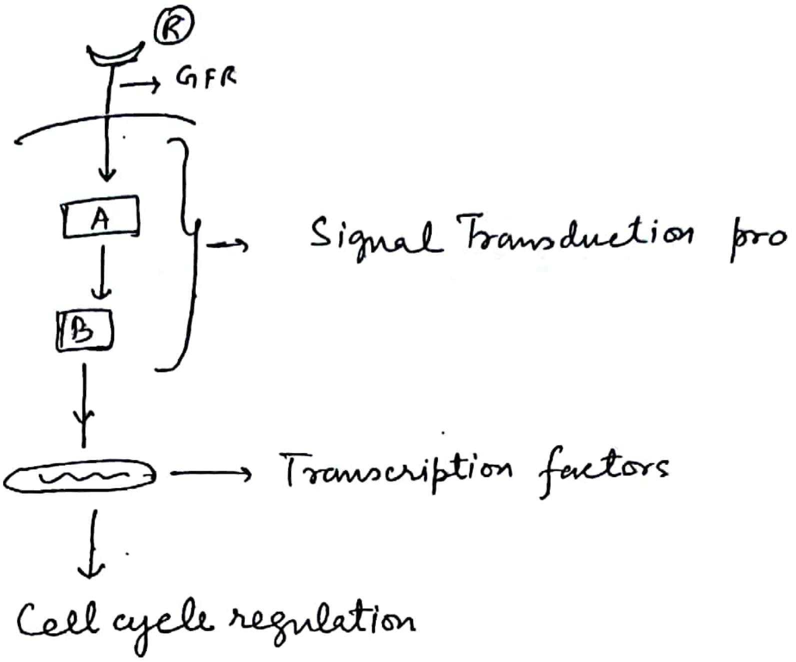
Neoplastic Schwann cells + Fibroblasts + Perineural like cells + Fibroblasts + Spindle cells (CD34 +ve) + Mast cells

S100 +ve

Heterogenous

NF1 associated

Wavy collagen & wavy buckled nuclei.



GFR

- EGFR (erbb1) Her2neu
- PDGFRA
- RET
- MET

STDPs

- KRAS
- BRAF
- PI3K

Tyrosine kinase Receptor TK

- ALK
- Ckit

Transcription f

PAX 8

myc

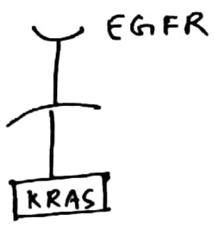
Cell cycle inhibitors CDKN2A/P16

Repair genes : BRCA1 & 2

p53

Rb

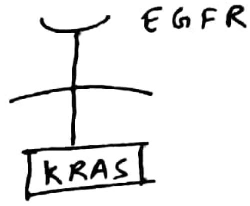
Lung



⇓

Mutually exclusive

Colon



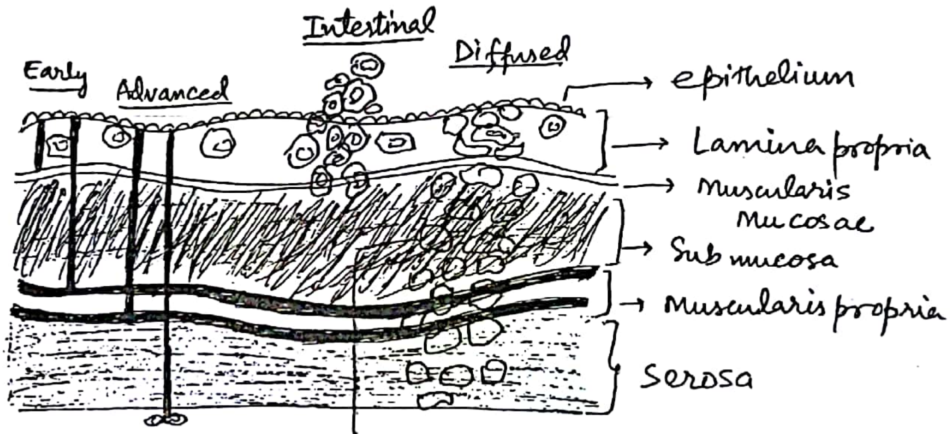
KRAS/NRAS mutation analysis is mandatory now before starting anti EGFR Therapy.

Q Mutation analysis of which of the following genes will not help in prognostication of colon cancer?

(A) EGFR
(B) KRAS
(C) P53
(D) MSH2
↓
DNA mismatch
↓
Ly Syndrome

Gastric Adenocarcinoma

- Depth of Invasion
  - Early
  - Advanced
- Histology
  - Diffuse
  - Intestinal



Signet Ring cell →  → Mucin

→ Nucleus pushed to the periphery

- No mucosal mass
- Loss of Rugal folds
- Transmural thickening - Infiltration → wall thickening

↓

LINITIS PLASTICA ← Leather bottle appearance

Disease	Effect on Intraluminal digestion	Terminal digestion	Transepithelial Transport	Lymphatic Transport
Celiac disease		⊕	⊕	
Lactase		⊕		
Whipples				⊕
Abeta			⊕	
Chronic Pancreatitis	⊕			

Hematology 23/04/18

Hematoncology

Hematopoietic Stem cells

→ Common Myeloid progenitor

→ Common Lymphoid progenitor → Lymphoblasts (B/T/Nk) → Lymphocytes (B/T/Nk)

Common Myeloid progenitor — DIFFERENTIATION →

- Erythroblasts → RBC
- Myeloblast → Granulocytes
- Monoblast → Monocytes
- Megakaryoblast → Platelets

(~~Differe~~ ~~Maturation~~ IMMATURATION)

Granulocytes → Neutrophils, Eosinophils, Basophils. (BEN)

Myeloblast → Promyelocyte → Myelocyte → Metamyelocyte → Band form → Neutrophil

Myeloblast: Prominent nucleoli

Promyelocyte: → Azurophilic primary granules

Neutrophil:
- Segmented Nucleus
- Increased no. of granules

While Maturation ~~RBCs~~
- Increased segmentation
- Increased functional granules
- Decreased size

Right Shift (Infection) ⇒ (↑ mature cells)

&

Left Shift (Hypoplastic marrow) ⇒ (↑ Immature cells) (Few lobes of Neutrophils Nucleus)

⇒ ARNETH Index or ARNETH COUNT

[redacted]

Leukemia ⟷ **Lymphoma**

Leukemia	Lymphoma
White in blood.	Lymphocytes ↓ Tumor
Liquid tumor	Solid tumor
Origin - Bone Marrow	Origin - Lymphoid organs → L.N, thymus, Spleen, tonsils. any where in body.
Malignant	Benign/Malignant.
Inv - Flowcytometry of peripheral blood or B.M. aspirate	Inv. ImmunoHistochemistry on Histopath sample. (IHC)

Gene Rearrangement

Change in location

- Translocation (Karyotyping, FISH — F In Situ Hyb.)
- Inversion

↓

gene 1 + gene 2

↓

mRNA (New fused) [FUSION TRANSCRIPT] } Inv. RT. PCR.

↓

New protein (oncoprotein)] IHC / Flow cytometry

↓

Cancer. (↑ Leukemias).

gene 1 → gene 2

↓

↑sed copies [(Amplification) → (FISH)]

↓

Increased protein → (IHC) [Overexpression]

↓

Increased unregulated proliferation

↓

Cancer. (↑ Lymphomas)

Mutation

- Change in sequence (PCR + Screening)
- → Commonly seen in Solid tumors.
- → Increased expression or Decreased expression ↓ New protein (IHC)

Hematolymphoid Neoplasms

- MYELOID
  - Acute Myeloid Leukemia (AML)
  - Myeloproliferative Neoplasms (MPN)
  - Myelo Dysplastic Syndrome (MDS)
  - MDS/MPN
- LYMPHOID
  - Hodgkin
    - Classical
    - Nodular lymphocyte Predominant
  - Non Hodgkins
    - B cell
    - T/NK cells

Acute Myeloid Leukemia (AML)

⇒ ↑se in Immature cells in marrow.

Origin :- Haematopoietic stem cells / progenitor

Marrow is taken over by bad cells → thus no place for good cells.

↓

- Anemia
- Thrombocytopenia
- Recurrent Infection

HSCs go to the extramedullary sites

↓

$\boxed{\text{Extramedullary Hematopoiesis}}$

↓ causing

$\boxed{\text{Spleenomegaly or Hepatomegaly}}$

⇒ Combination of Maturation arrest + Clonal proliferation

⇓

Increased Immature cells.

FAB classification

AML - with minimal differentiation (M0)
AML - without maturation (M1)
AML - with maturation. (M2)
AML - with promyelocytic maturation (M3)
AML - with myelomonocytic differentiation. (M4)
AML - with monocytic differentiation (M5)
AML - with erythroid differentiation (M6)
AML - with megakaryotic differentiation. (M7)
↳ (↑ risk of Myelofibrosis)

Indentated nucleus ⇒ Monocyte

2017 WHO AML Classification.

≥20% Blastcells in Marrow. (<20% - MDS)

1. AML with recurrent genetic abnormalities.

Do not require 20% Blast counts for diagnosis (exceptions)
- AML with t(8;21)
- AML with t(15;17) $\boxed{M_3}$
- AML with t(16;16) or inv(16)

} Favourable Prognosis

AML with t(6;9)
AML with 11q23 rearrangements (MLL gene)
} Unfavourable Prognosis

AML with t(9;22) (WHO 2017)

Phil. Chromosome, 22nd chr. formed after translocation

AML with normal Karyotype.
- (i) Biallelic $\boxed{\text{CEBPA}}$ mutation
- (ii) $\boxed{\text{NPM}}$ mutation

(2017) } Favourable prognosis

8;21
15;17
16;16
6;9
11q23
9;22

2. AML, therapy related.

Alkylating Agents
Epipodophyllotoxins

3. AML, with dysplasia related changes.
   - With prior MDS
   - Without prior MDS

4. AML, Not otherwise Specified (NOS)
   Previous FAB M0 – M_2, M_4 – M_7. (30-40%) More common in adults.

AML with $\boxed{t(8;21)}$

$\boxed{RUNX_1T_1}$ $\boxed{RUNX_1}$ } → Important for myeloid & lymphoid differentiations & maturation.

- with M_2 Morphology.

AML with $\boxed{t(15;17)}$

PML (Promyelocytic Leukemia) RARA (Retinoic acid receptor α)

PML-RARA Fusion

Myeloblast ⟶ Promyelocyte ⟶ Myelocyte

PML-RARA fusion blocks the maturation

Thus ↑ Promyelocyte

Atypical promyelocyte + Stacks of Auer rods ⇒ $\boxed{\text{FAGGOT Cells}}$

↑ Granules → Procoagulate in nature
↓
[Widespread thrombosis → [Disseminated Intravascular Coagulation] DIC
↓
[Consumption of all platelets & coagulation factors.
→ Consumption Coagulopathy.
↓
Bleeding

All Trans Retinoic Acid (ATRA) Remove the block!
Arsenic

[AML $\bar{c}$ DIC = M3
[AML $\bar{c}$ best prognosis = M3

AML with t(16; 16) or Inv(16)

Gene - Core binding factor β (CBF β)

Usually shows $\boxed{M_4}$ & $\boxed{M_5}$ morphology.

→ Nuclear creases/folds/Indentation.

Monoblast (NSE +ve)

✓ M_4 = MPO + NSE +ve.   Myeloblast ⇒ Auer Rods

✓ M_5 = NSE +ve

Blasts
- Agranular → MPO +ve.
  - Few Auer Rods → M2
  - Many Auer Rods → M3
- Agranular (MPO-ve)
  - NSE+ve → Monoblasts → M5
  - PAS +ve (Block like PAS Positive)
    - Cytoplasmic Vacuoles → M6
    - cytoplasmic blebs → M7
    - TdT +ve → Lympho blasts
- Few Auer Rods + Monoblasts → M4 (MPO +ve, NSE +ve)

TdT — Terminal deoxynucleotidyl Transferase

MYELOPROLIFERATIVE NEOPLASMS (MPNs)

Origin - Hematopoietic cells (stem cells)/Progenitor cells

PANMYELOSIS → ALL, Myeloid cells with (!) GRANULOCYTE DOMINANCE

⇓

Chronic Myeloid Leukemia (CML)

t (9; 22) = fusion of BCR - ABL 1 (→ Tyrosine kinase) on Chr 22 ⇓ Philadelphia Chr.

⇒ Variable excess of BCR may fuse with a fixed sequence of ABL1

(i) Major BCR (M-BCR) + ABL 1 = 210 KD = P210 → CML

(ii) Minor BCR (m-BCR) + ABL1 = 190 KD = P190 → ALL

⇓

Unregulated Tyrosine Kinase

⇓

Using TYROSINE KINASE

(!) AML can have both (p210 & p190) WHO 2017

In **CML** chronic phase

TLC ↑ → Differential c. ⇒ Marrow preferable over blood

- ✓ Myelocyte peak
- Neutrophil peak
- Blasts < 2%
- ✓ Basophilia
- Eosinophilia +/-

} ✓ Garden Party Appearance / College Girl Appearance

Additional cytogenetic changes

⇓

Progression to Accelerated Phase

↑ TLC → Differential c.

Blasts 10–19% (marrow > blood)
Basophilia > 20% (blood)

Thrombocytosis / Thrombocytopenia
↑ing Spleenomegaly.

Unresponsiveness to Tyrosine Kinase Inhibitors (WHO 2017)

⇓

Additional Genetic Changes.

⇓

Blast Crisis

Blasts ≥ 20% (Marrow > Blood) OR Extra medullary Blast Proliferation (Skin, CNS)

75% (Myeloid) Blasts — 25% (Lymphoid) Blasts.

RT PCR → To monitor response to treatment. & to followup

⇒ Neutrophil Alkaline Phosphatase (NAP)
(Leukocyte ALP)

NAP → Number – ↑ses (Leukemoid reaction)
NAP → Function – ↓ses (CML)

2. Erythrocyte Dominance :- Polycythemia vera

[redacted]

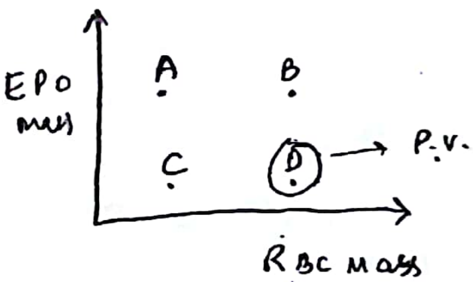
→ Epo (erythropoietin)
Epo receptor
→ JAK2 (JANUS KINASE) (Non receptor kinase Tyrosine kinase)
→ Nucleus ↓ Erythrocyte formation & proliferation.

Unrequired & unregulated Erythropoiesis
↓
- RBC Mass ↑
- Hb > 16.5 g/dl (M)
  > 16.0 g/dl (F)
- Hematocrit
  > 49% = (M)
  > 48% = (F)

Polycythemia vera, Essential thr., Myelofibrosis → JAK 2 → Activating Point Mutation
⇓
Proto oncogene → Oncogene → Unrequired & unregulated Erythropoiesis → POLYCYTHEMIA VERA

Biopsy – Erythroid hyperplasia [redacted]
⇓
At 617th position
Valine → Phenylalanine

- EPO Level = Normal/Subnormal



↑ Risk of thrombosis ⇒ Budd Chiari Syndrome

Platelet function defects may be present → Bleeding manifestation

(3) Essential Thrombocytosis / Thrombocythemia
(Megakaryocyte dominance)

Platelet count $> 450 \times 10^9/L$

Biopsy ⇒ Mature enlarged Megakaryocytes

Genes - JAK2 (40%)
MPL } (NHD 2017)
CALR }

↑ Thrombosis (+)

→ ~~Epo~~ Thrombopoietin
→ Thrombopoietin Receptor
JAK2
↓
Thrombopoiesis

JAK2 → Activating point mutation (JAK2 V617F) 40%
⇓
Essential Thrombocytosis.

Bone Marrow Aspirate & Biopsy indicated

(4) Primary Myelofibrosis

Denovo Marrow Collagen deposition

Panmyelosis (JAK2, MPL, CALR) ⟶ Atypical Megakaryocytes
↓
TGF β } ⇒ Fibrosis
PDGF }

↓ cellularity in marrow
⇒ Extra Medullary Hematopoiesis (EMH)

- Tear Drop cytes/cells / Dacrocytes (distorted RBCs)

⟶ 0 0

- Precursors of RBCs & Granulocytes.

Erythroblasts } LeucoErythroBlastic Reaction (LEBR) ⇒ Myelofibrosis
Leucoblasts }

Biopsy is diagnostic
↳ Aspirate is Dry tap

Demonstrate fibrosis (Pathways)
↳ Reticulin Stain (Silver → Black)
↓
Visualize Reticulate fibers
(Fibrosis of liver)

AML c̄ Myelofibrosis

Biopsy indicated

Myelofibrosis secondary to some inciting events

↓

<u>Myelophthisic Anemia.</u>

→ Metastatic Carcinoma

→ Storage disorders

→ Granulomatous Inflammation

→ Radiotherapy / Chemotherapy.

<u>MYELODYSPLASTIC SYNDROME</u>

Disordered growth <u>in marrow</u> ⟶ Cytopenia in blood.

At least 10% of the cells of a series should have dysplasia to call it significant.

MDS can occur in a single lineage ⇒ Single Lineage Dysplasia (MDS – SL)

or Multi lineage ⇒ (MDS – ML)

MDS with ring sideroblasts

MDS with excess blasts.

	Blood cytopenia	Bone marrow dysplasia	Ringed Sideroblasts	Blasts in marrow.
MDS-SL	Uni/Bi cytopenia	Uni cell line	<15%	<5% (No AUER ROD cells)
MDS-ML	UNI TO Pancytopenia	More than one cell line	<15%	<5% (No A.R. cells)
MDS-RS Ring Sideroblasts SL	Uni cytopenia	One or two cell lines	>15%	<5 8% (NO A.R. cells)
MDS-RS MLD	Uni to Pan cytopenia	More than one cell line	>15%	<5% (NO A.R. cells
MDS-EB1 Excess blasts	Uni to Pan cytopenia	One to three cells lines	None or any	5-9% (NO A.R. cells)
✓ MDS-EB2	Uni to Pan cytopenia	One to three cell lines	None or any	10-19% Auer Rods

Erythroid Dysplasia

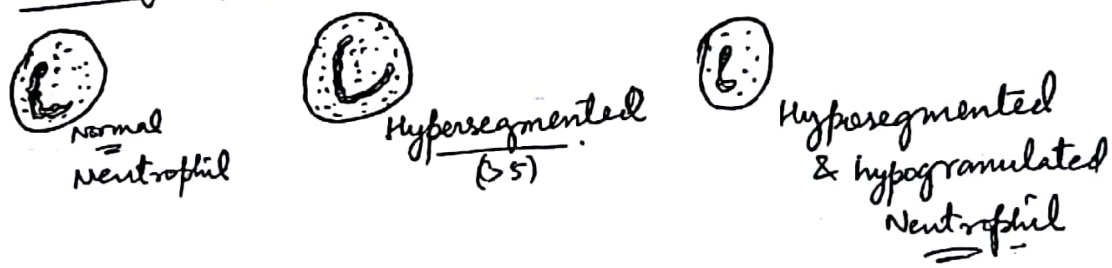
Perlic Stain


→ Prussian blue deposits
[Iron ladden Mitochondria
around the nucleus)

⇓

Ring sideroblasts

Granulocyte Dysplasia



⇓

Pelger Huet Anomaly :-
- Inherited condition
- Neutrophils are morphologically abnormal but functionally normal.

Pseudo Pelger Huet anomaly

PINCE NEZ Appearance

Megakaryocyte Dysplasia

	Dysplastic
Large cell →	Small cell
Single nucleus →	Multinucleated
Multilobed →	Hyper/Hypolobulated

⇒ Most significant Dysplasia for diagnosis of MDS.


→ Pawn ball megakaryocytes - small megakaryocytes c̄ multinucleation & Hypolobation

Most common cytogenetic abnormality in MDS
In Adults
In Children
Overall
} ⇒ $\boxed{\text{Monosomy 7}}$ (who 2017)

MDS/MPN

Chronic Myelomonocytic Leukemia (CMML) — $>1 \times 10^9/L$ ē monocytosis $\geq 10\%$ of all Leucocytes

Myelo ⇒ Dysplasia; monocytic ⇒ Proliferation

Juvenile Myelomonocytic Leukemia (JMML)

Atypical Chronic Myeloid Leukemia (ACML) (Philadelphia Chr. −ve)

New confirmed addition by WHO.
↳ MDS/MPN with ring sideroblasts and thrombocytosis

Lymphoid Neoplasms

HODGKIN LYMPHOMA	NON HODGKIN LYMPHOMA
– Involves contiguous group of lymph nodes. Contiguous → (sharing common border)	Non contiguous group of lymph Nodes
– Almost always Nodal (Axial L.N.)	Can be Nodal / Extranodal (Walders Ring, Mesentric LN) commonly affected
– Bimodal age distribution (young adults & old)	No specific age group.
✓ Neoplastic cells <<< Non neoplastic cells (1:10 – 1:100)	Almost all cells are neoplastic.

HL ⇒ Classical R.S cells
Lacunar cells
Mononuclear cell
L&H cell.

NHL

- B cell (more common) ✓
  - Precursor → B-acute Lymphoblastic Leukemia/Lymphoma
  - Peripheral/mature
- T/NK Cell
  - Precursor → T/NK-Acute Lymphoblastic Leukemia/Lymphoma
  - Peripheral/Mature

Acute lymphoblastic Leukemia / Lymphoma

Most common Cancer in children

B-ALL
- Children
- Leukemia ($\uparrow$ Tendency)

T-ALL
- Adults
- Lymphoma ($\uparrow$ Tendency)
- Thymic mass → SVC obstruction

Also seen with
- small cell CA.
- Hodgkin Lymphoma

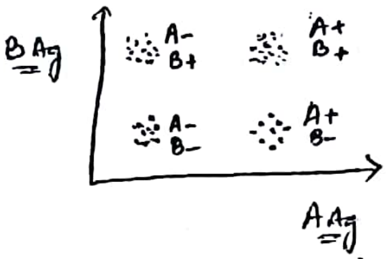
Lymphoblast
- Large cell
- $\uparrow$ N:C ratio
- Less/no prominent Nucleoli
- Scanty cytoplasm
- Agranular
- Block like PAS +vity.

Prognosis of ALL

Unfavourable	Favourable
<1yr, >10yrs	2 - 9 years
CNS involvement Testicular involvement	Absent
Male.	Female
T. phenotype	B phenotype
t (9;22) (P190)	t 12;21 RUNX1
11q23 MLL gene	
Hypoploidy (<45 chr)	Hyperploidy (>47 chr.)

Priority for ALL prognosis
① – Response to treatment (steroids)
② – Cytogenetics – Polidy / Translocations
③ – Clinical features.

Flowcytometry :→



Myeloid Markers :–

~~Myelo~~peroxidase (MPO) :– Lineage Specific marker for granulocytes

CD 13, CD 33] :→ Seen in granulocytes + Monocytes (Myeloid Lineage marker)

CD 11c, CD 14, CD 64] :→ Monocytic markers.

CD 71, CD 235] :→ Erythrocytic markers

CD 41, CD 61] :→ Megakaryocytic markers.

Lymphoid Markers :→

T cell

- CD 1a → Thymocytes, Langerhan cells
- CD 2 → Almost all T cells
- CD 3 ⟶ Part of T receptor (more diagnostic)
- CD 4 ⟶ Helper T cell.
- [illegible] T [illegible] & Subset of B cells

$\boxed{\text{Pan T cells: } 2, 3, 7}$

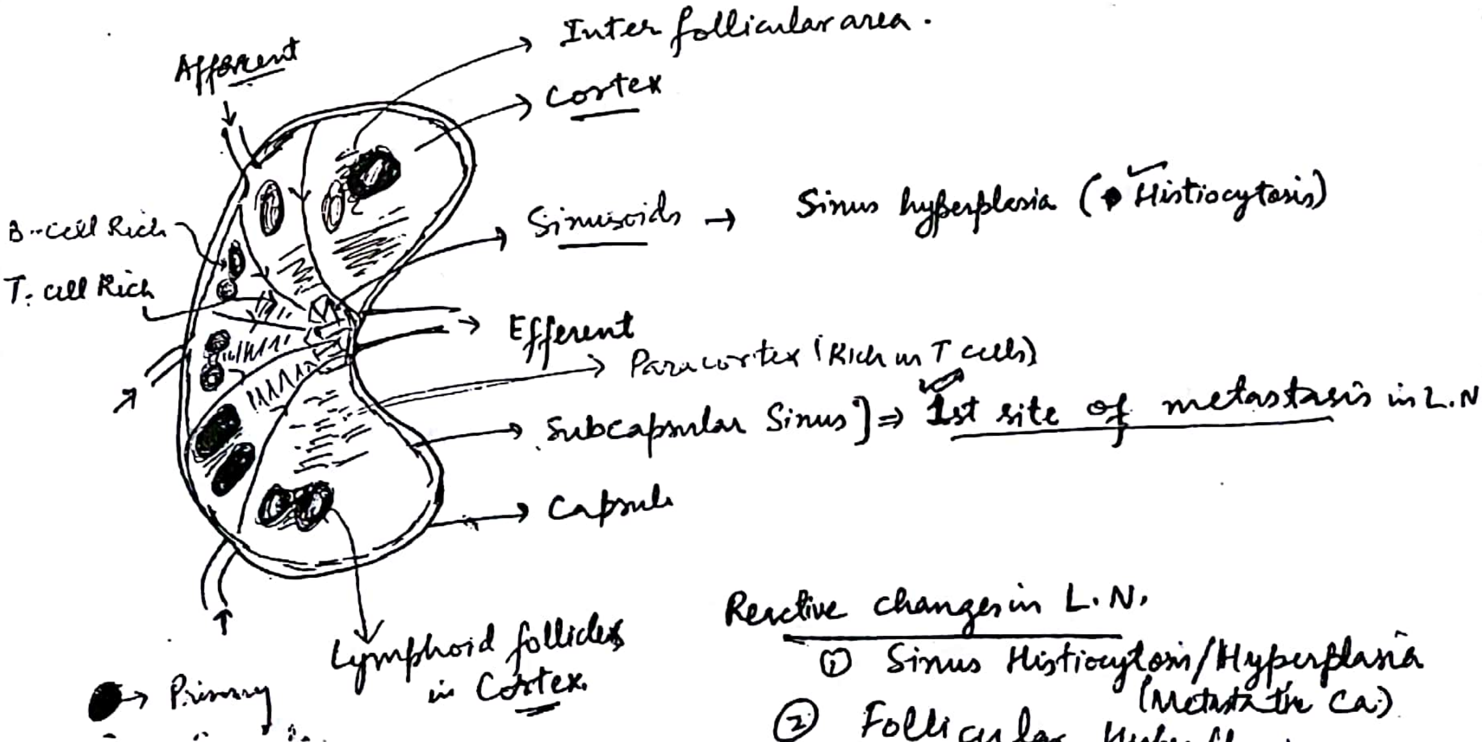
B. cell markers

- ✓CD19 → Linerage specific and consistently expressed.
- CD 20 → Marker of choice for B cell lymphomas (Mature)
- ✓CD 21 → EBV receptor
- CD 23 → Activated B cells
- CD 79A → Almost all B cells
- ✓PAX 5 (New marker) → B cell trancription factor. (PGI)

[♡ → First love of B cell: PAX 5]

Miscellaneous markers

- ✓CD 34:- Hematopoietic stem cells + Progenitors
- HLA DR: Blasts (immaturity)
- CD10: Precursor B cell + T cells
  Germinal centre - B cells
- CD15 - Mature granulocytes



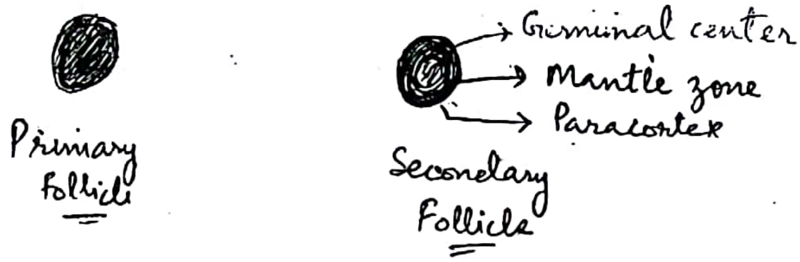
Reactive changes in L.N.

1. Sinus Histiocytosis/Hyperplasia (Metastatic Ca)
2. Follicular Hyperplasia

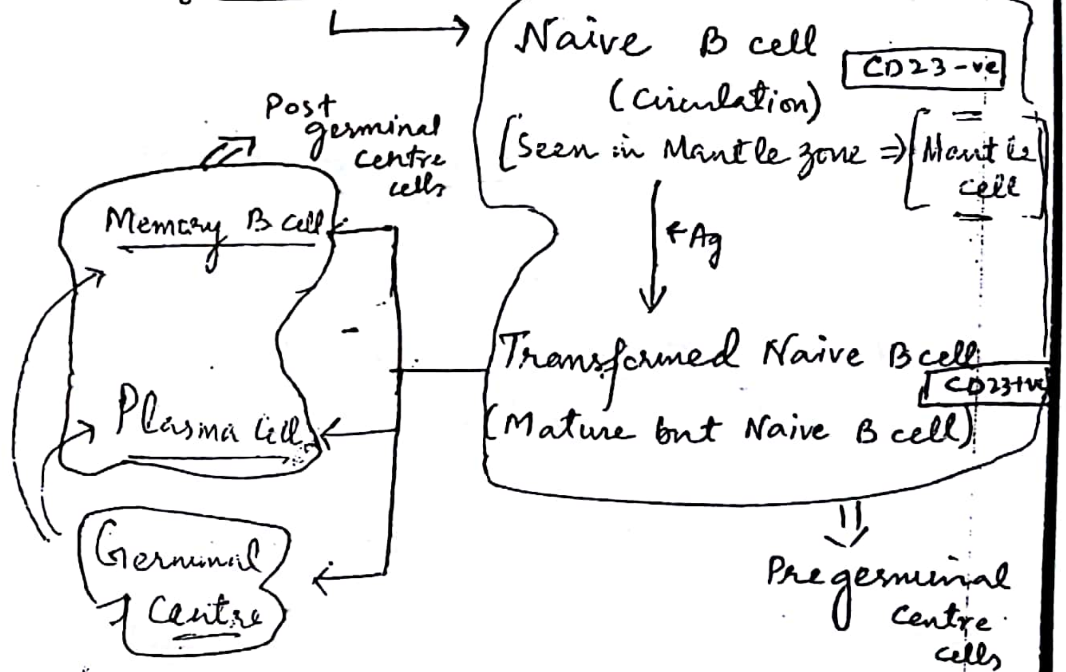
Follicular hyperplasia – Humoral ~~[illegible]~~ immunity requirement ↑↑

Paracortical hyperplasia – ↑ T cell mediated immunity
↳ infected B cells but increases T cells.

⇒ Plasma cells and Histiocytes are found in Medulla.



B Lymphoblast in Marrow



Pre Germinal centre	Germinal centre	Post Germinal Centre
→ Mantle cell lymphoma $\boxed{\text{CD23 -ve}}$	Burkitt lymphoma	Marginal zone lymphoma
→ Chronic lymphocytic lymphoma/Leukemia. $\boxed{\text{CD23 +ve}}$	Follicular lymphoma	Plasma cell neoplasm
	Diffused Large B cell lymphoma (DLBCL)	Lymphoplasmacytic Lymphomas (LPL)
		CLL
		DLBCL
$\boxed{\text{CD5 +ve / CD10 -ve}}$	$\boxed{\text{CD10 +ve / CD5 -ve}}$	$\boxed{\text{CD5 -ve / CD10 -ve}}$

① Mantle cell lymphoma

Elderly

Nodal & Extranodal

↓

GIT - Polypoidal appearance.

May be high grade. (Small cells c̄ cleaved Nucleus)

$\boxed{t(11;14)}$

→ IgH (Heavy chain Ig) (Promoter)

→ Cyclin D1 (cell cycle regulator which pushes the cell cycle forward)

↓

Overexpressed (IHC)

New marker for cyclin D1 Negative MCL

→ [redacted] May AIIMS17

SOX 11

IHC

- CD20 +ve
- CD79a +ve
- CD5 +ve
- CD23 -ve
- CD10 -ve
- Cyclin D1 +ve

(2) Chronic Lymphocytic Leukemia / Small Lymphocytic Lymphoma (CLL/SLL)

Small cell — Resembling small lymphocyte
Non cleaved
Low grade


→ Coarse/clumped cromatin
→ Scanty cytoplasm.

Soccer ball appearance 

- CLL is most common Leukemia in adults
- Almost exclusively in the elderly!

Peripheral Blood :- Uniform monotonous population
[Convent School Girl appearance]

Tumor cells have very less Vimentin ⇒ Intermediate filament of cytoplasm
↓
Become fragile
↓
Making a smear ~~[illegible]~~ disrupt easily.
↓
 → Smudge cells / Basket cells

Lymph Node Histology

Diffused effacement of LN architecture (No demarcation between cortex & medulla seen) Most common Pattern

Pro lymphocyte (another type of tumor cell)

 (Rapidly multiplying cell)
→ enough cytoplasm.
thus lighter staining.

 → Proliferation centres (Pseudo follicular pattern) → Most Pathognomonic pattern of CLL

More the proliferation centres Poor the prognosis (AIIMS 2022)

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Most common cytogenetic Change

50% – Deletion 13q

20% – Trisomy 12 (47 XY +12)

New marker for CLL – $\boxed{\text{LEF1}}$

	MCL	CLL
Nuclear	+	+
CD5	+	+
CD23	~~+~~ –	+
CD20	+	+
cyclin	+	–

Best marker to differentiate b/w MCL & CLL → CD23

③ Follicular Lymphoma

Nodal >> Extranodal

Most common NHL in western hemisphere

Dual population.

Multiple peripheral Nucleoli

Centroblast

Cleaved Nuclei

Centrocyte

WHO grading of FL = No. of centroblasts per high power field.

gr I < 5

gr II 6-15

gr III > 15

t (14; 18)
↓ IgH (Promoter)
↳ BCL2 (anti apoptotic) ⇓ over expression (IHC)

⇒ Back to back closely packed follicles with very little interfollicular space.

To differentiate from follicular hyperplasia
↳ $\boxed{\text{BCL 2}}$ is the marker of choice
⇓
+ve in germinal centres of follicular lymphoma & -ve in germinal centre of follicular hyperplasia

BCL II is normally expressed in mantle zone.

IHC
- CD 20 +
- CD 79a +
- CD5 -
- CD10 +
- BCL6 (germinal center) +
- BCL2 +

(4) Burkitt Lymphoma

✓ Extranodal >>> Nodal

✓ - EBV +ve

Types
- Endemic (African continent) — Most common site is Jaw
- Sporadic — Most common site Ileocaecum
- Immunodeficiency associated (HIV) — Most common site - CNS, GIT

✓ 8 = B
↓
Chr. No 8.
"C myc" ✓ Transcription factor

⇒ t(8; 14)
↳ IgH ⇒
- Kappa (K) Chr 2 t(2; 8)
- Lambda (L) Chr 22 (t(8;22))

To assess proliferation of tumor cells – marker used is

Ki-67 // (MiB-1)

(% of cells express)

Lymphomas

< 40% – low grade

> 40% – High grade

[different cut offs for different tumors]

Ki 67 in Burkitt lymphoma ≃ 100%

i.e., rapidly proliferating & rapidly dying cells

High Mitotic Index ⟹

Starry sky appearance

Also seen in

- ALL
- BLBCL
- Variants of MCL
- Reactive lymph nodes.

IHC

- CD 20 – +ve
- CD 79a +ve
- CD 5 –ve
- CD 10 +ve
- BCL6 +ve
- BCL2 –ve
- Translocation Cmyc +ve

New marker is [illegible] WHO 2017

⑤ DL BCL (Diffused Large B Cell Lymphoma)

Diffused → Diffused effacement of architecture

Large → Nuclei of tumor cell is 3x – 5x that of normal lymphocyte nucleus

Most common NHL worldwide

↳ Molecularly heterogenous.

BCL6 rearrangement / BCL2 rearrangement / Cmyc rearrangement | Frequency

* Most common Ca arising in HIV patients //

DLBCL is high grade lymphoma.

Large cell

Histological variants
- Centroblastic
- Immunoblastic
- Plasmoblastic → Most common in Immunodeficiency.

IHC
- CD 20 +ve
- CD 79a +ve
- CD 5 -ve
- CD 10 +ve
- C myc +/-
- BCL2 +/-
- BCL6 +ve.

Ki 67 > 40% (not as high as Burkitt)

Low grade Small cell Lymphoma } $\xrightarrow{\text{Can transform}}$ High grade Large cell Lymphoma

⇓

Richter transformation

(6) Marginal Zone Lymphoma

⇒ Diagnosis of exclusion
(CD5 - , CD 10 -) (CD43 +ve)

NODAL & Extranodal ⇒ MALToma
⇓
Chronic inflammatory states
- Sjogren's Syndrome
- Hashimoto thyroiditis
- H. Pylori Gastritis (regression of MALToma on treating H.Pylori)

New marker for Marginal zone lymphoma
⇓
IRTA 1
MNDA.

MALT (Mucosa-associated lymphatic tissue)

(7) Lymphoplasmacytic Lymphomas

Commonly associated with Hep C.

↓ Lympocytes

↓ Plasma cells

↓ IgM ⟶ forming pentamers

↓ ↑ Viscosity in blood

⇓ Waldenstorm's Macroglobulinemia (WM)

[redacted]

JIPMER 2017

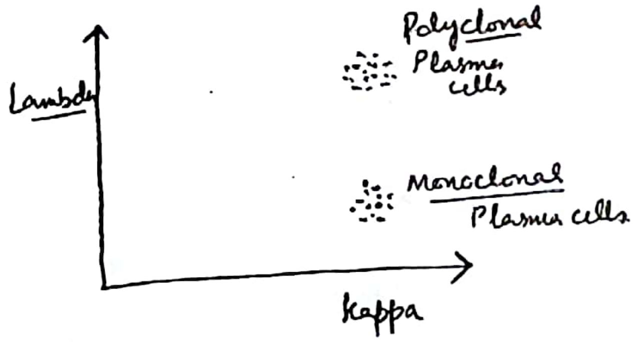
LPL in CNS c̄ WM

⇓ Bing Neel Syndrome

(8) Plasma Cell Neoplasms

Clonal plasma cells ⟶ { Abnormal Immunoglobulins + Excess of Light chains }

↓ Monoclonal Proteins (M-Proteins)



Deletion 13q

Monoclonal Gammopathy of Uncertain Significance (MGUS)

Heavy chain disease

↓ Rare monoclonal gammopathy occurs in malnurished population

A/K/A ↳ Mediterranean Lymphoma.

Smoldering Myeloma

Plasma cell Myeloma

	Monoclonal Gammopathy of Uncertain Significance (MGUS)	Smoldering Myeloma	Plasma cell Myeloma
M-protein (g/dl)	< 3	> 3	> 3
Bone marrow clonal plasma cell (%)	< 10%	> 10%	> 10%
Symptoms	Absent	Absent	Present

MGUS → Premalignant condition.

MGUS

↙ Ig M-MGUS (MYD88 mutation) ↓ Lympoplasmacytic lymphoma progression

↘ IgG/IgA-MGUS ↓ Plasma cell myeloma progression

(MYD88 L265P) Mutation.

Plasma cell Myeloma

Tumor plasma cells

↙ Abnormal Igs + Excess light chains

Recurrent infection ⇒ ⇓ M/c/c Death in M.M.

⇓

⇒ Excreated in Urine as Bence Jones protein

⇒ Light chain Nephropathy ⇓ 2nd M/c/c of R.F.

→ Renal Insufficiency & ⇓ ↓ erythropoietin ⇒ Anemia (Multifatorial) (Normocytic Normo chronic)

2nd M/c/c death in MM

↘ Osteoclast Activating Factors

↓ [redacted]

↙ Stimulates Osteoclasts ⇓ [redacted]

↘ Inhibits Osteoblasts ↓ ALP levels (N/↓)

Bone resorption

- Hypercalcemia
  - ↓ Most common Cause of Renal failure in Multiple Myeloma.
- Blanched out Lytic bone lesions [redacted]

CRAB Symptoms of M.M.
- Hypercalcemia
- Renal insufficiency
- Anemia
- Bone Resorption/lesion

Tumor plasma cells

IL-6 → Survival, proliferation & level correlate with disease activity.

Symptoms of P.C. Myeloma

>20% of Plasma cells in peripheral blood

⇓

Plasma cell leukemia

How to Diagnose Multiple Myeloma As Per 2014 IMWG Criteria

1st → Look for >10% clonal plasma cells in bone marrow or biopsy proven plasmocytoma (either bony or extramedullary)

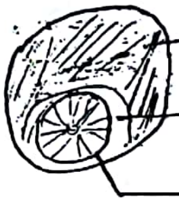
2nd ⇒ Any of the myeloma defining events.

End Organ Damage (CRAB)
- Hypercalcemia
- Renal Insufficiency
- Anemia
- Bone lesions

Any one or more of malignancy biomarkers
1) >60% of plasma cells in Bone marrow.
2) Involved/uninvolved Serum free light chain ratio ≥100
3) >1 focal lesion on MRI (>5 mm)

In Multiple myeloma order of frequency of Ig secreted

⇒ Ig G (50%).
Ig A (20%)
Ig D
Ig E
Ig M



Globular inclusions
⇓
Mott cell/Grape cell

Intra Nuclear inclusion
⇓
Dutcher Body
common

Intra cytoplasmic
⇓
Russell Body

Malignant plasma cells are ⇒ Binucleate/ Multinucleate
⇒ Have Inclusions

Most common cytogenetic abnormality associated is
Rearrangements of $\boxed{14q (IgH)}$

Most common gene involved is $\boxed{\text{cyclin D1}}$

Drugs used ⇒ Ibrutinib
Rituximab.

(9) Mature T cell lymphomas

1. Mycoses Fungoides (Most common cutaneous T cell Lymphoma)

↓

Leukemia when developed is called [redacted] (Tumefaction of Skin Lesions)

CD4+

⇒ Cerebriform Nuclei

SEZARY LUTZNER cell → Invade epidermis & form [redacted]

(10) Adult T cell Lymphoma/Leukemia

Caused by HTLV-1

→ Flower shaped/ cloverleaf shaped Nuclei

(11) Anaplastic Large cell Lymphoma

T cell lymphoma

ALK positive

↳ Anaplastic Lymphoma Kinase (Receptor tyrosine kinase)

Rearranged in

- ALCL
- Lung Adenocarcinoma
- Inflammatory Myofibroblastic tumor AIIMS

Mutated in

- Neuroblastoma
- Hereditary hemorrhagic telengiectasia II (JIPMER)

"Hallmark cells"

Embryoid Nuclei | Horseshoe Shaped Nuclei | R.S like cells

HODGKIN Lymphoma

- Involues Contiguous group of LN.
- Almost always Nodal —— Most common is cervical group. (~~Neck~~/Axial)
- Bimodal age
- Neoplastic cells << Nonneoplastic cells

ASSOCIATED WITH EBV

RS cells + Mixed infl. cells

Classical (CHL) [EBV+ve]

Nodular Lymphocyte Predominant HL (NLPHL) → [EBV-ve] → Best prognosis of all HL

Classical (CHL) subtypes:

Nodular sclerosis	Mixed cellularity	Lymphocyte depleted	Lymphocyte Rich
M = F	M > F	M > F	
Mediastinal Lymphnodes		Strongest Association c̄ EBV & HIV	
Most common subtype overall	Most common Subtype in India	Worst Prognosis	
Least associated c̄ EBV (5-10%)	Most common in HIV		
Best prognosis in Classical HL	Most common in EBV HL		

CD15, CD30 — Classical marker for HL.

→ Large/gaint cell
Binucleate
Prominent macronucleoli

Owl's eye appearance

origin - germinal centre B cells
Size - 45 μm

<u>Variants</u> –  Mononuclear cell / Hodgkin cell.

 – Lacunar cell → Nodular Sclerosis

 – Popcorn cell / LP cell / L & H cell
⇓
NLPHL

	RS cell	LP cell
CD45 (Leucocyte common Ag)	–	+
CD20	–	+
CD10	–	+
BCL6	–	+
CD30	+ 100%	–
CD15	+ 75-85%	–
PAX 5	Weakly +ve 95%	Strongly +ve

Hairy cell Leukemia

- Tumor cells resemble the features of memory B cells
- Mutation has been identified in the hematopoietic stem cells.

↓

[redacted] V600E ↳ Gouth position Val → Glutamate

↓

Signal transduction protein

Also seen in - Langerhan cell histiocytosis
- Pilocytic Astrocytoma
- Papillary Carcinoma Thyroid
- Malignant Melanoma

BRAF inhibitor

↳ VEMURAFINIB


Hairy cells (frayed borders)

In marrow → Individual cell fibrosis


→ Fried egg appearance → also seen in Oligodendroma, Mycoplasma c.

Aspirate - Dry Tap
Biopsy - Diagnostic

- Also infiltrates Spleen → Spleenomegaly (involves Red pulp)
- Pancytopenia
- Tumor cells stain positive for acid phosphatase
  ↓
  wash them with tartarate
  ↓
  Stain is retained.

⇐ Tartarate Resistant Acid Phosphatase (TRAP) +ve.

Associations
- Monocytopenia ⇒ ↑sed Risk of Infection with atypical Mycobacterium
- Erythema nodosum
- Polyarthritis Nodosa

IHC - $\boxed{\text{Annexin A1}}$ most specific marker

CD 11c
CD 103
CD 25

Langerhan Cell Histiocytosis

- Tumor of Immature Dendritic cells (Langerhan's cells)
- [redacted] mutation
- Clinically

Unisystem

- **Unifocal**
  - Eosinophilic granuloma
- **Multifocal**
  - Calvarial bone defects
  - Exophthalmos
  - Diabetes Insipidus
  - → Hand Schuller Christian Triad

Multisystem

- Cutaneous
- Hepatic
- Spleenic
- Pulmonary
- → Letterer Siwe Disease

Histology

Coffee Bean Nuclei (Longitudinal groove)

→ Langerhan cells

→ Eosinophill.

Also found in
- Granulosa cell tumor
- Brenner tumor
- Papillary Ca. Thyroid
- Chondroblastoma

Electron Microscopy

⇒ Most specific marker ⇒ LANGERIN (CD 207)

Pentalaminar crystalline structures with fusiform dialation (Tennis Racquet appearance) ⇒ Birbeck Granules

Pulmonary LCH

- Neoplastic
  ↓
  BRAF V600E
- Reactive
  ↓
  Smoking
  ↓
  Cessation of smoking causes regression of disease

IH

CD 1a

$\boxed{\text{S-100}}$ → Neuroectodermal marker

HLA-DR

- Malignant Melanoma
- Schwannoma
- Lipoma / Liposarcoma
- Sustentacular cells of pheochromocytoma.

RBC Disorders

Anemia

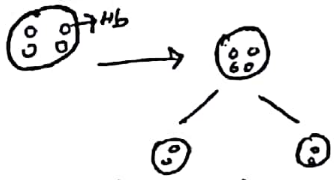
Means Absence → Blood

↓sed O_2 carrying capacity
Reflected on ↓ed Hb

- Too little production
  - Aplastic anemia
  - Nutritional deficiency
- Too much Destruction
  - Hemolytic anemia
- Blood loss.
  - Acute
  - Chronic

Hemolytic Anemia

Microcytic anemia
⇓
Defective in formation of Hb.



Microcytosis

Protoporphyrin → Heme (Fe)
Sideroblastic anemia

Fe ↓
- Iron deficiency
- Anemia of Chronic disease

Heme ↓ Globin - Thalassemia
Hemoglobin

Reticulocyte :- Immature RBC with retained RNA due to improper enucleation & maturation

→ Polychromatophilic cell seen as routine peripheral smear
→ Purplish due to bluish staining of RNA.

On Supravital Stain → Reticulocytes meshwork/ reticulum of blue deposits of RNA

Routine - Polychromatophilic
Supravital : Reticulocyte
↳ New Methylene blue
→ Brilliant cresyl blue
→ Crystal violet

Reticulocytosis → marrow is normally functioning
→ Marrow is in hurry.

Normal Retic count = 0.5 - 1.5%

Hemolytic Anemia

↑sed RBC destruction

~~Extrinsica~~

Intrinsically wrong c̄ RBC
↓
or There is Intracorpuscular defect.
⇓
Extravascular Hemolysis
↳ Spleenomegaly.

Extrinsically wrong c̄ RBC
↓
or There is Extracorpuscular defect.
⇓
Intravascular Hemolysis

Lab findings

Signs of ↑sed defect/destruction
- Hemoglobinemia/uria
- ↑sed Indirect Bilirubin
- ↑sed LDH
- ↓sed Haptoglobulin

Signs of compensatory production
- Reticulocytosis
- Bone marrow hyperplasia

Hereditary Spherocytosis

Defect in membrane proteins

Integral

Almost never involved in H.S → Glycophorin (CD235)

Band3 (most abundant Integral glycoprotein)

Peripheral

⇒ Ankyrin - Most common def.

⇒ Spectrin - most severe def.

Responsible for biconcave shape of RBC

⇒ Protein 4.1

⇒ Protein 4.2 (Palladin)

Most common inheritence is Autosomal Dominant.

Loss of Surface area → Spherical Shape ⇒ Splenic macrophagic destruction of RBC formed

↓ Mean corpuscular Hb Conc. (MCHC)

Less deformable

⇒ Osmotically ~~morphologically~~ more fragile.

⇒ RBCs without any central pallor

↳ Characteristic feature of Spherocytes

#

Paroxysmal Nocturnal Hemoglobinuria (PNH)

- Paroxysmal → Episodic
- Nocturnal → Decreased pH in body during night activates complement.
- Hemoglobinuria → Intravascular hemolysis

⇓

Acquired genetic defect

PIGA gene

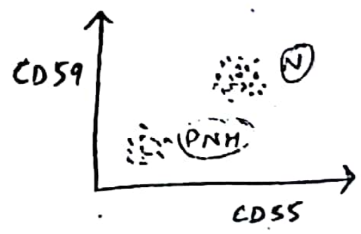
GPI anchored proteins

⇓

(Protect cells from complement mediated damage)

GPI anchored Proteins

- Membrane Inhibitor of Reactive lysis (MIRL) (CD59) ← Most common & Important
- ~~Reactiv~~ Decay Accelerating Factor (DAF) (CD55)
- C8 binding protein



Triad of
- → Hemolytic anemia
- \# → Pancytopenia
- \# → Thrombosis (50%) - common cause of Death. (Multifactorial)

5-10% ⇒ Progress to AML/MDS

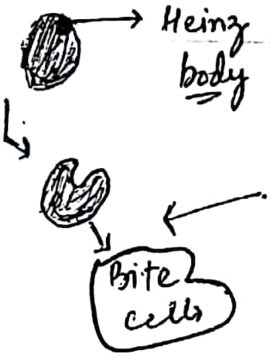
Stem cell Transplant ⇓ Treated.

Diagnosis
- Ham's test
- Sucrose lysis test
- Flowcytometry ⇒ Best technique

G6PD Deficiency

HMP Shunt → Detoxify H_2O_2

In G6PD deficiency → when there is oxidative damage
↓
H_2O_2 is not detoxified
↓
Free radical damage to Sulfhydryl groups of Hb.
↓
Denaturation of Hb
↓
HEINZ Body
↓
Can be visualized on Supravital Stain



→ Denatured Hb does'nt take other stains.

When stained Routinely

Heinz body → capable of damaging the membrane
↓
EV Hemolysis

without membrane damage - IV Hemolysis

G6PD deficiency → X linked recessive
Males full expression
Females - Carriers.

Oxidant damage

Infections
Drugs — Antimalarials, Dapsone, Cotrimoxazole, Nitrofurantoin
Fava beans

CDNA

Adult Hb

HbA	$\alpha_2\beta_2$	96%
HbA_2	$\alpha_2\delta_2$	3%
Hb F	$\alpha_2\gamma_2$	1%

⇒ α is coded by a pair of genes on chr.16

⇒ β is coded by a single gene on chr.11.

Sickle cell Anemia

Glutamate changes to valine at 6th position of β globin chain

↓

Hb becomes sticky.

In reduced state → the stickiness of Hb causes polymerization

↓

HbS polymers distort the shape of RBC

Sickle cell

Drepanocyte

- Fragile
  ↓
  Hemolysis
- Can cause microvascular occlusion
  ↓
  Hypoxia → Ischemia
  ↓
  Tissue damage.

If there is repeated occlusion in spleenic tissue,

↓

repeated cycles of damage & fibrosis

↓

Autosplenectomy → ↑ed risk of infection by capsulated organism

Autosomal Recessive

ββ – (N)

$\beta^s\beta \rightarrow \alpha_2\beta_2 = \boxed{HbA}$ → Interferes $\bar{c}$ polymerization of HbS.

$\boxed{\alpha_2\beta^S}$ ← Heterozygous

↳ Sickle cell trait

$\beta^S\beta^S \Rightarrow \alpha_2\beta^S_2$ – $\boxed{HbS}$ Homozygous condition

Diagnosis

Hb Electrophoresis]– Qualitative

High performance liquid chromatography

[HPLC]

↳ Quantitative & Qualitative

Sickling test

(Na Meta bisulphite) is used.

or (Na dithionate)

$\boxed{\text{THALASSEMIA}}$

α or β thalasemia (α↓ or β↓)

↓ globin production → Microcytic cells

If $\alpha\downarrow \Rightarrow \beta\uparrow, \delta\uparrow, \gamma\uparrow$

$\beta\downarrow \Rightarrow \alpha\uparrow$

Thus getting Excess unpaired chains

↓

form tetramers

↓

$\beta\uparrow \Rightarrow \beta_4$ – HbH (Golf ball Inclusions) Precipitate ⇒ Hemolysis

$\gamma\uparrow \Rightarrow \gamma_4$ – $\boxed{\text{Hb Bart's}}$

α thalasemia

Most common Genetic Change ⇒ Deletions

αα | αα → (N)

αα | α– → 1/4 deletion / silent carriers

αα | – – → 2/4 deletion

or α thal-trait

α– | α–

α– | – – → 3/4 HbH disease

– – | – – → 4/4 deletion → Hydrops fetalis

β thalasemia

Clinically — Major / Intermedia / Minor

β - Genetic changes

β - Ⓝ

$β^+$ - Reduced production of β] ⇒ Splicing Mutations

$β^0$ - Absent production of β] ⇒ Chain Termination mutation

When both alleles are mutated
↳ Homozygous (Major)

When only one allele is mutated
↳ Heterozygous (Minor)

Major	Minor
- Severe anemia	- Mild to moderate anemia
- Dependence on blood transfusion.	- No history of blood transfusion
Target cell +ve Microcytic cell	- Target cell +ve Microcytic cell
⇒ Considerable amount of anisocytosis & reticulocytosis	⇒ Uniformly microcytic so no anisocytosis

Target cell OR CODOCYTE

Microcytic cell.

Anisocytosis ⇒ Variation of SIZE
↓
Absence

Poikilocytosis ⇒ Variation of Shape

RDW - Red cell Distribution Width

Range over which RBCs are distributed.
↳ Anisocytosis Confirmation by RDW.

β thal. Minor

At least on allele is functional (β)

Mild reduction in β

↓

Mild excess of α → binds ē δ

⇓

$\alpha_2 \delta_2$ → HbA_2 ↑

β thal major

Both alleles are affected

Severe reduction in β

Severe excess of α → binds more ē γ → HbF ↑

& δ also → HbA_2 ↑

HPLC - Quick Interaction

	HbA	HbA_2	HbF	Additional.
Normal	~96%	< 3.5%	<1%	—
β thal. Trait	< 96%	3.5-9%	Normal or ↑sed	—
β thal Major	~3%	Variable	>85%	—
Sickle cell Trait	40-60%	~3.5%	Normal or Slightly ↑sed	HbS-30-40%
Sickle cell Homozygous	Markedly Reduced	~~Variable~~ Slightly ↑ or Normal	5-25%	HbS - 70-90%

Autoimmune Hemolytic Anemia (AIHA)

↓

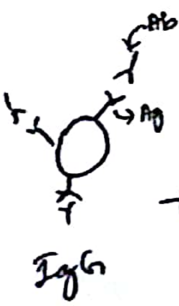
Auto antibodies targetted against RBC Ags

- Warm Antibody type (IgG Antibodies Active at 37°C) (No clumping in PB Smear)
  - Primary
  - Secondary
    - Autoimmune disorder ⇒ SLE
    - Drugs
    - Lymphoid Neoplasms ⇒ CLL
- ~~Cold~~ Cold Agglutinin Type (IgM antibodies active below 37°C)
  - Acute (mycoplasmal Infection) (Infectious mononucleosis)
  - Chronic (Clumping is found in PBS)
    - Idiopathic
    - Lymphoid neoplasms.
- Cold Hemolysin Type (IgG antibody active below 37°C)
  → Donath Landsteiner Ab.
  Rare; Occurs mainly in children following viral inf.

As RBC is covered with antibodies

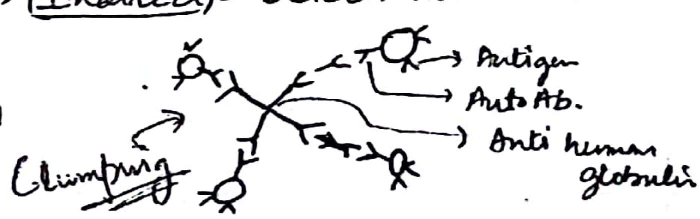
- Compliment mediated membrane damage
  - → IVH
  - → Spherocytes ⇓ Most common cause is AIHA
- Spleenic macrophages destroy them ↓ EVH

Coomb's Test
- → Direct – Detects Abs bound to RBCs.
- → Indirect – Detects Abs in serum



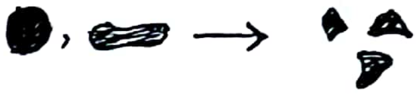
\+ Anti Human globulin

(Antibodies against Auto antibodies) ⟹



Microangiopathic Hemolytic Anemia (MAHA)

↓ Small ↓ Vessel pathology



- In Vasculitis (SLE)
- DIC
- TTP/HUS

| Thrombi formation

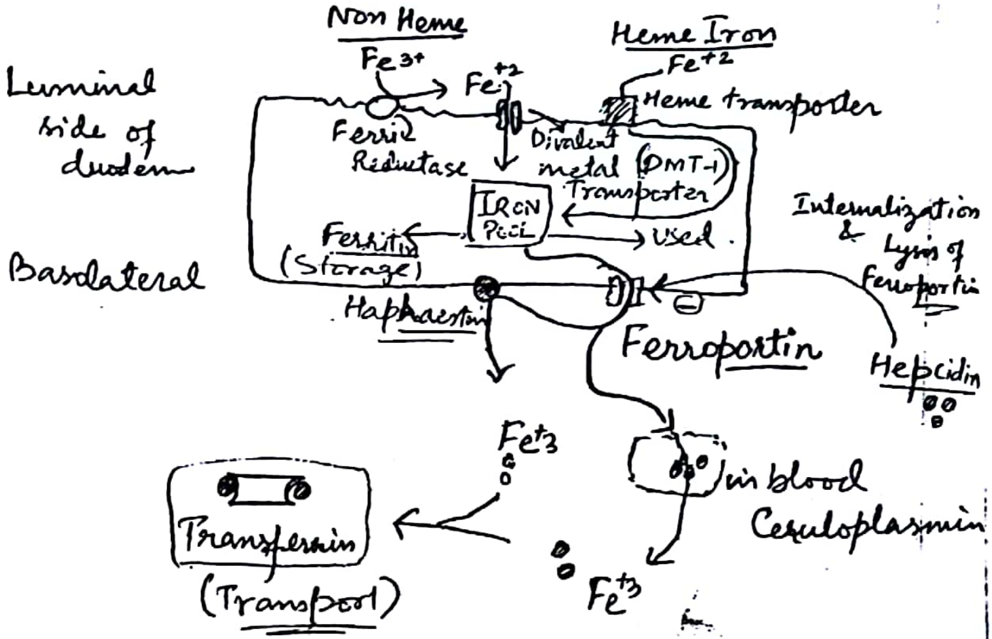
- Schistocytes
or
Schizocytes
or
Fragmented RBCs
or
Helmet Shaped

Macroangiopathic Hemolytic Anemia

⇓

Prosthetic valves
Coarctation of Aorta

Iron Absorption



Plasma Ferritin
↓
Reflects Iron Stores

Cu deficiency
↓
Can also lead to Iron deficiency
As Haphaestin is ⇒

$\boxed{\text{Hepcidin}}$ ↳ ↓sing the exit of Iron from cell to blood.

$\boxed{\text{TIBC}}$ ⇒ Reflection of Transferrin Production in body.

Transferin % Saturation

Iron deficiency Anemia

- Earliest smear finding is : $\boxed{\text{Anisocytosis (RDW↑)}}$
- After treatment first response: $\boxed{\text{Improvement of Mood}}$
  Sign is Reticulocytosis
  (↑se in Reticulocyte Hb)
- Pencil Shape cells +ve ↳ characteristic of Iron def. anemia

Megaloblastic Anemia :-

- It is a macrocytic anemia, but every macrocytic anemia is not a megaloblastic anemia
- Macrocytic anemia ⇒ $\boxed{MCV > 100}$

Folic acid / Vit B_{12} ↓ ⟶ Retarded DNA Synthesis

↓

Immature Nuclei
+
Mature cytoplasm
+
Big cells

Megaloblasts in marrow
↓
Macroovalocytes in blood
↓
Earliest finding

other cells are also affected
↓
Giant metamyelocytes
+
Hypersegmented Neutrophils
↓
most characteristic

Ineffective erythropoiesis → Improperly formed cells
↓
Destroyed in marrow itself
↓
Hemolytic phase
↓
Jaundice

Platelets

Injury to vessel
↓
Vasoconstriction
↓
Primary Platelet ~~Plaque~~ Plug
(unstable) → Coagulation Cascade.

↑

Fibrin clot

Primary Hemostasis	Secondary Hemostasis
- Disorders will present c̄ Superficial hemorrhages	Deep haemorrhages
- Not associated with family history	Family history +ve
Thrombocytopenia	Hemophilia (A/B/C)
Immune Thrombocytopenic Purpura (ITP)	Clotting factor inhibitors
Thrombotic Thrombocytopenic Purpura (TTP)	

Von Willebrand Disease

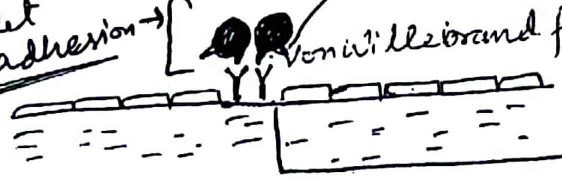
Disseminated Intra Vascular Coagulation.

Platelets release ① granules
- α
- δ — Serotonin, ADP, Ca^{+}

② Thromboxin A_2

Fibrinogen → Coagulation → Fibrin
(on the surface of primary platelet plug)

gpIb/IX (CD42) → CD42a, CD42b] ⇒ Deficiency ⇓ Bernard Soulier Syndrome

Platelet adhesion → [ von Willebrand factor →] von Willebrand disease

Exposure of Subendothelial Collagen

↓ Platelet activation

→ ADP, Calcium, Adrenaline [Degranulation]

Platelet aggregation

→ gpIIb/IIIa (CD41) (fibrinogen receptor) ↓ Glanzmann Thrombasthenia

→ Fibrinogen

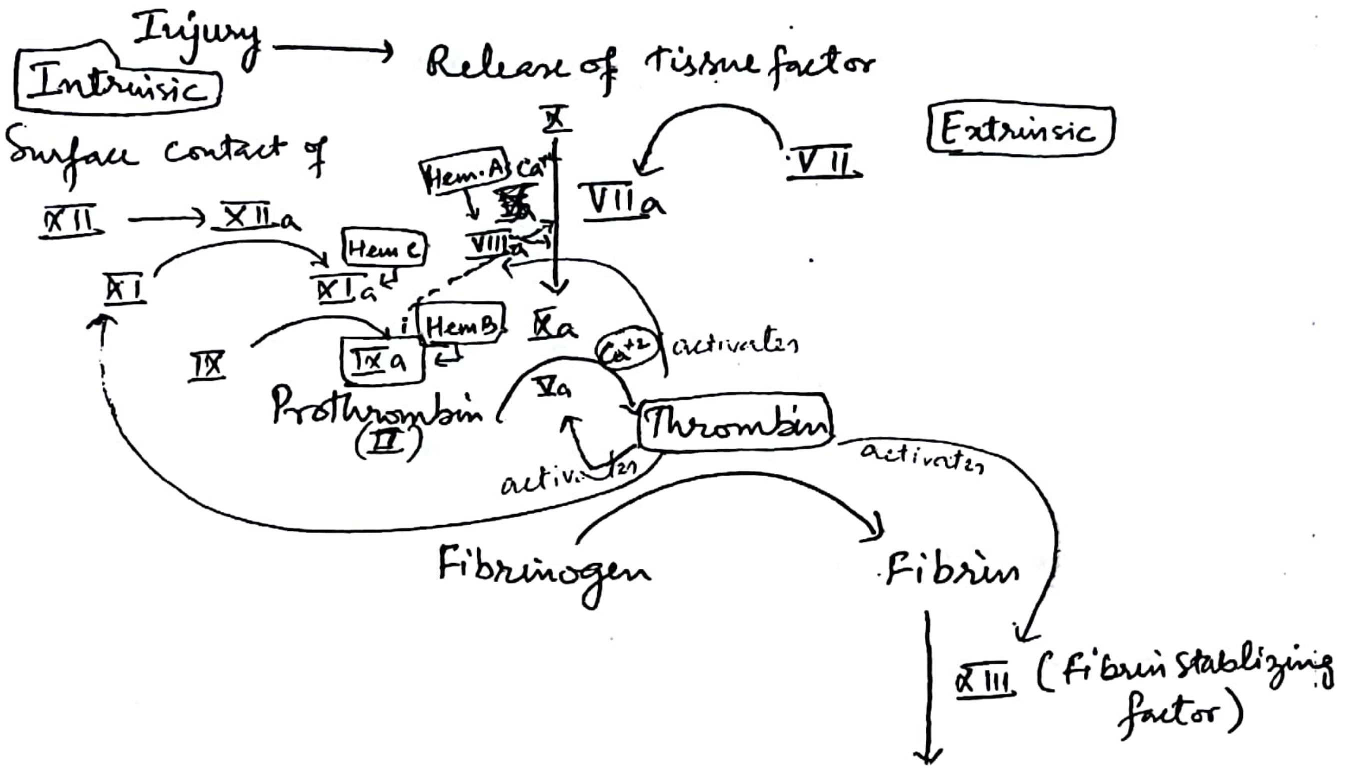
Umbilical Stump bleeding

Platelet function Aggregometry (PFA)

Agonists are given to stimulate Primary ~~plaque~~ Plug formation

- Ristocetin → Plt. ~~aggreg~~ adhesion
→ If no curve/flat curve
↓
VWF or gpIb/IX ↓↓
↓
VWD or gpIb/IX def.
↓
BSS

Injury to vessel
↓
Release of Tissue factor



Polymerises & Stablises the CLOT.

XII def → Asymptomatic

Intrinsic Pathway

Measured by APTT (Activated Partial Thromboplastin Time)
[25 - 40 sec (N)]

Extrinsic Pathway

Measured by PT.
[11 - 14 sec (N)]

Factor VII - K dependent factor

Prothrombin time is measured as
Platelet Poor Plasm + Tissue factor [Thromboplastin]
↓
Phospholipids (Platelets are missing)
↓
Calcium (Monitor stopwatch)

APTT

Platelet poor plasma + Surface Activator
(glass, silica, kaolin)

↓

Phospholipids (Platelets are missing)

↓

Calcium → (Start stopwatch)

Immune Thrombocytopenic Purpura

Auto antibodies against Plt. Ags.

Most common — $\boxed{\text{gp IIb/IIIa}}$

Destruction of plts. by spleenic macrophages [TOC – Splectomy]

Children (Acute) – Following Infection

Adults (chronic) – No H/o Infection
Auto immune disorders.

Marrow → Megakaryocyte Hyperplasia
(not required of Dx)
as it is non specific

VWF (von Willebrand Factor)

Produced as large multimers $\xrightarrow{\text{ADAM TS 13 enzyme}}$ broken in Small molecules

↓

Act as carrier for factor VIIIa

Deficiency of ADAM TS 13 enzyme

↓

Large multimers of VWF will cause widespread platelet ~~aggre~~ adhesion followed by aggregation.

⇓

Thrombotic Thrombocytopenic Purpura

If VWF is deficient ⟶ No primary plug formation
↓ If severe ↳ APTT ↑
↓ Bleeding

Functional deficiency of VIII
will lead to ↑ APTT in VWD.

DIC

Procoagulant molecules (cancers, Autoimmune)
Extensive endothelial injury

⇓

Wide spread thrombosis → PT ↑, APTT ↑] due to Coagulation factor consumption.
→ Bleeding

Also called as Thrombohaemorrhagic Disorder.

Bleeding Pt. with APTT ↑
↓
Mixing Studies) ⇒ Pt plasma + Normal pooled plasma
50:50
↓
Repeat APTT

- APTT normalize
  (Confirms Coagulation factor deficiency)
  ↓
  Factor Assay.
- APTT is still ↑sed
  ↓
  Confirms presence of Clotting factor Inhibitors
  ↓
  Lupus Anticoagulant → Dilute Russel Viper venom Test

Inherited Thrombophilia

Thrombin activates Protein C
Protein S

Cleavage site for APC

Normal → Va
VIIIa

$\xleftarrow{\text{degradation}}$ Activated Protein C (APC)

Thrombin → Protein C → Activated Protein C (APC)

Normal → **Leiden Mutation** → (no cleavage site)

Thus APC resistance

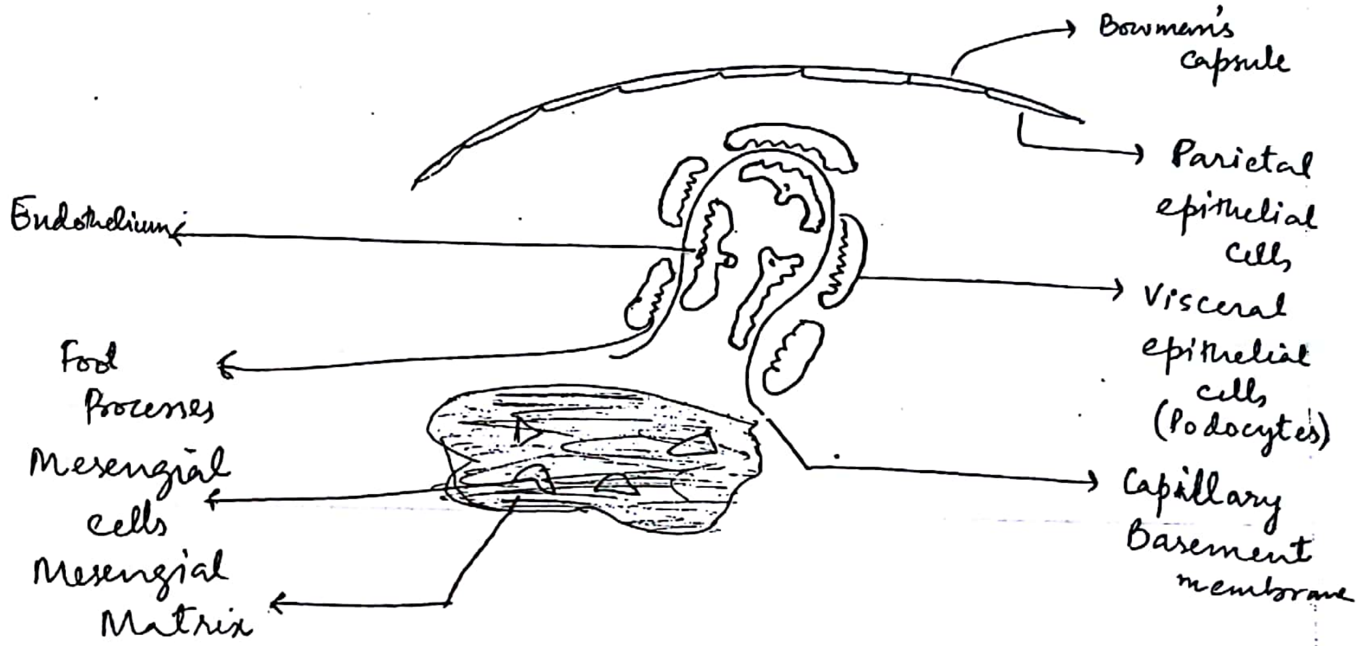
Intrinsic

Extrinsic

(2)

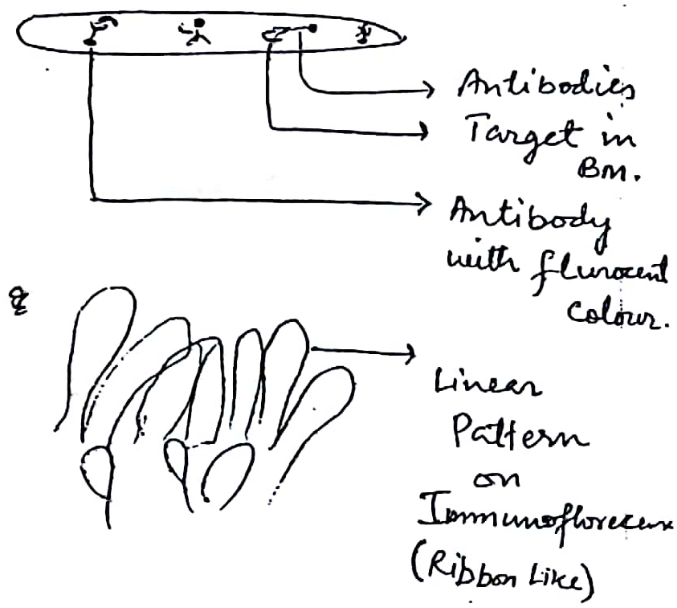
Renal Pathology

Ultrastructure of Glomerulus



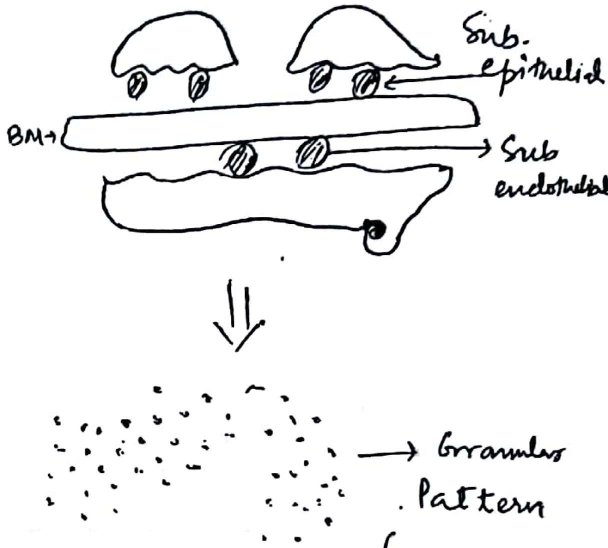
Mechanisms of Glomerular Injury

① Antibodies to components of Basement membrane.



2) Immune Complex deposition

Circulating | Insitu



3) ANCA mediated.

(Anti Neutrophil cytoplasmic Antibody)

⇓

(No pattern on immunoflorecense.)

Clinically

Hematuria - Damage to glomerular basement membrane (most often due to inflammation)

↓

Nephritic

Proteinuria - Derangement of capillary wall + Podocytes.

Nephrotic :→ Massive Proteinuria

[> 3.5 g/day : Nephrotic Range]

Massive Proteinuria → Loss of antithrombin → Thrombotic Tendency

Massive Proteinuria → Hypoproteinemia

Hypoproteinemia → Liver tries to compensate by ↑ing protein synthesis ⇒ ↑ Lipoproteins ⇒ Hypolipidemia + Lipiduria

Hypoproteinemia → ↓ Pressure in vessels → Loss of fluid → Edema

Loss of fluid → ↓ GFR → RAS + → Salt & H_2O Retention → Edema

Edema → Periorbital

Edema → Generalized

Salt & H_2O Retention: Most important Cause of Edema in Nephrotic Syndrome.

Clinical

Proteinuria

Lipiduria

Hypolipidemia

Hypoproteinemia

Clinicopathological features

1. Minimal change disease (Nephrotic)
2. Focal Segmental glomerulosclerosis (Nephrotic +/_ Nephritic)
3. Membranous Nephropathy (Nephrotic)
4. Membroproleferative glomerulonephritis (Nephrotic + Nephritic)
5. IgA Nephropathy. (Nephritic)
   ↳ (BERGER's Disease)
   (BUERGER ✗ → Thrombangitis Obliterans)
6. Post infectious glomerulonephritis (Nephritic)
7. Rapidly progressive glomerulonephritis (RPGN) (Nephritic)

Pathological Assessments

① Light Microscopy
- H & E
- Special Stains → PAS (Magenta)
  → Jone's Methanamine Silver (Black)

② Immunofluoresence → No pattern.
→ Linear
→ Granular
→ Anyother

③ Electron microscopy — Site of deposition
— Associated changes

Minimal Change Disease

(NIL LESION or LIPID NEPHROSIS)

? Cytokine mediated podocyte Injury.

↳ Dramatic Response to steroids.

↳ This condition is associated with Hodgkin lymphoma. & Tcell lymphomas

LM ⇒ None

IF ⇒ None

$\boxed{EM}$ ⇒

Effacement/Loss of foot processes

Focal Segmental glomerulosclerosis

Focal ↓ Only some of the glomeruli are involved

Segmental ↓ Only a part of glomerulus is involved

Sclerosis ↓ Thickening/Hardening ↓ Hyaline deposition

Eosinophilic Amorphous Acellular

Homogenous glassy.

[Intracellular/Extracellular]

Tiny Tiny damage to BM ⟶ Tiny Tiny leakage of plasma proteins

↓

Podocyte damage ⟶ Dying Podocyte $\xrightarrow{\text{Debris}}$ Deposited outside the capillary wall (Hyalinosis)

↓

loss of foot processes.

\+

Lipid Deposition.

Obliteration of Capillaries and sclerosis of glomerulus

⇒ Idiopathic (Primary)

⇒ Secondary focal segmental glomerulosclerosis

→ Virus : HIV / HepB / Parvovirus

→ Hypersensitivity Nephropathy

⇒ Reflux Nephropathy.

Cholesterol emboli

⇒ Drugs: Heroin/ analgesic/ Pamidronate.

⇒ Oligomeganephronia

⇒ Renal dysgenesis

⇒ Alport's syndrome

⇒ Sickle cell disease

⇒ Lymphoma

⇒ Radiation nephritis

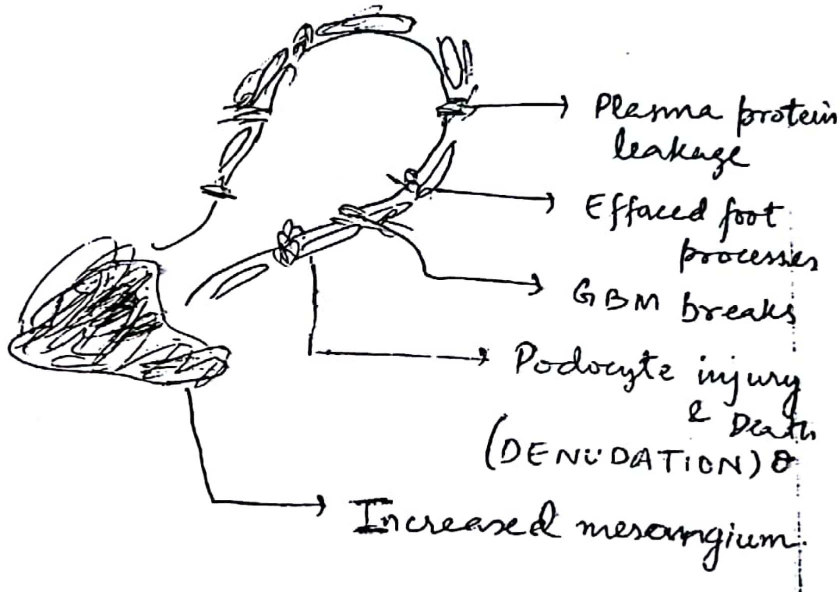
⇒ Familial podocytopathies

Gene	Protein
NPHS 1 mutation	nephrin
NPHS 2 mutation	podocin

LM:

Mesangial widening by sclerosis
↓
Obliterate capillaries
↳ Hyaline nodules.

IF: Usually no pattern > Granular.

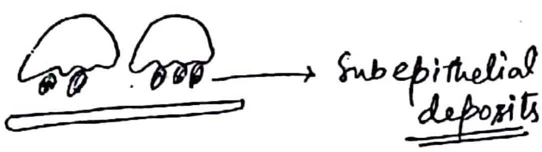


Membranous Glomerulopathy

Primary

Antibodies against

Phospholipase A_2 Receptors on podocytes



Secondary

1. Drugs – Gold, NSAIDs, Penicillamine
2. Infection – Hep B – Hep C – HIV – Leprosy; Syphilis
3. Cancers – Lung – Colon – Melanoma
4. Autoimmune – Hashimoto & SLE

LM :-
- Thickened basement membrane H & E
-

IF

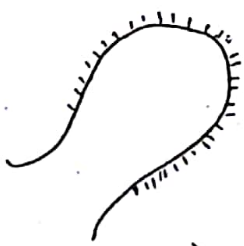
Granular IF.

EM

New Basement membrane material deposition (due to Irritation)

Subepithelial deposits.

⇓



Spike and Dome Appearance

- EM
- Silver stain (best viewed)
- PAS

Membrano proliferative Glomerulonephritis

- Membrano → Thickened BM → ⊖ Subendothelial Deposits
- proliferative → ↑sed cellularity (↑ cells) → ⊖ Mesangial & Endothelial (Reactive)
- Glomerulonephritis → Inflammation → ⊖ GBM damage & Podocyte Damage.

Type I (Most common)

Type II (Dense deposit disease)

Type III (Now removed)

Type I

Subacute bacterial endocarditis

Systemic Lupus erythematous

Hepatitis C ± cryoglobulinemia

Mixed cryoglobulinemia

Hepatitis B

Cancer: Lung, breast & Ovary (germinal)

Type II

C_3 Nephritic factor-associated

Type III

Complement Receptor deficiency

LM

Hyper-cellular glomerulus.
- Mesangial cells ↑ed (more)
- Endothelial cells ↑ed

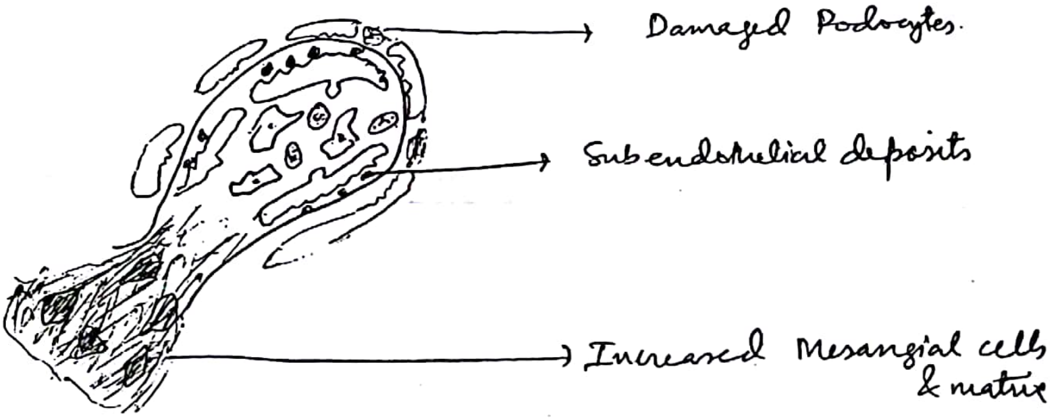
AKA Mesangio capillary glomerulonephritis

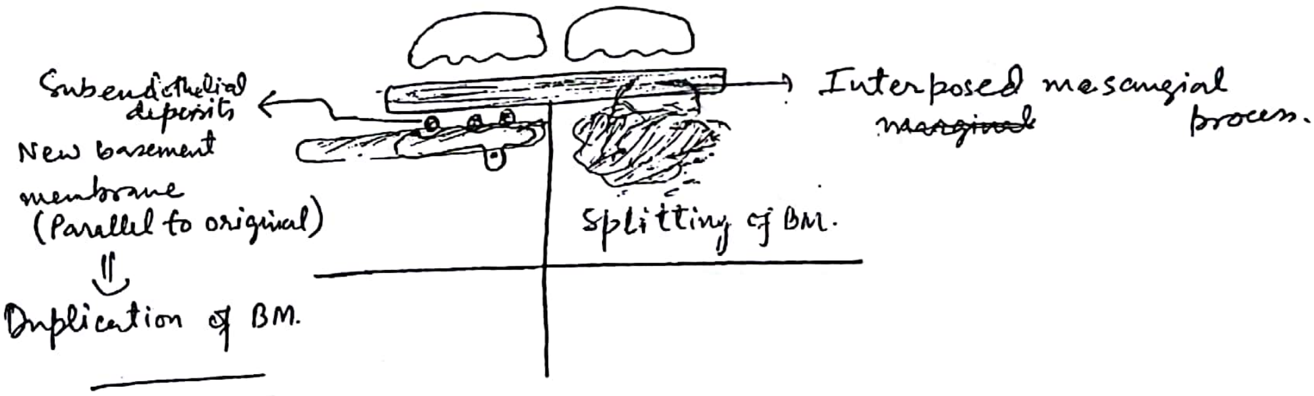
Inflammatory cells ⊕

Silver

IF - Granular.

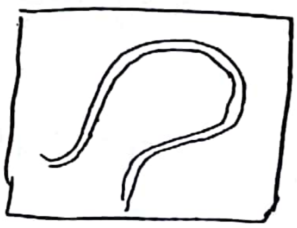
EM





⇓

TRAM TRACK APPEARANCE
or
DOUBLE CONTOURING



IgA Nephropathy (BERGER'S Disease)

↑sed IgA : Following Respiratory/GIT infections.
< 1 week Q.

↓sed IgA clearance Liver diseases
(↓ed hepato billary clearance)

Abnormal IgA : Celiac disease Q

Also attacks anchoring filaments in hemidesmosome of Dermal papillae thus leading to
⇓
Dermatitis herpetiformis.

LM
Mesangial widening.
(even on PAS)

IF : Mesangial pattern.

EM :

⇓
Mesangio proliferative
(Mesangial widening)

PSGN

POST INFECTIOUS

After 1-3 wks of throat infection or 3-5 weeks of Skin Infection

(POST) INFECTIVE → Streptococcal
Staphylococcal
Viruses
Parasites

or

(Acute) Proliferative → Increase in number of cells.
↓
Hypercellular glomerulus.

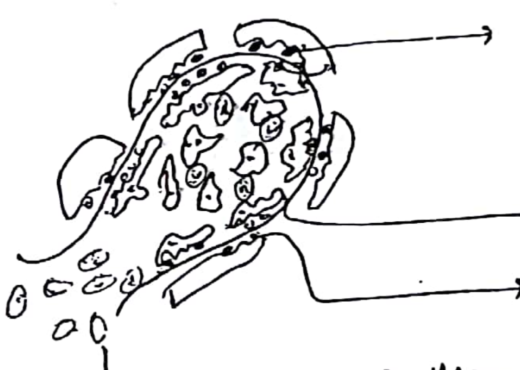
Glomerulonephritis
↓
Inflamation
↳ Damages GBM

Onset is quick:
transient
Rarely leads to chronic renal failure.

LM:- Hypercellular glomerulus
increased Endothelial cells — More ↑ (Max seen)
Mesangial cells
Inflammatory cells

IF : Granular

EM



→ Subepithelial deposits (MC & Characteristic)

→ Endothelial ↑

→ Subendothelial (early)

→ Mesangial cells ↑

Most common cause of Nephrotic Syndrome.

In adults – Focal segmental Glomerulosclerosis

In Children – Minimal change Disease

In Elderly – Membranous nephropathy.

Most common cause of Glomerulonephritis.

Primary GN in world ; IgA Nephropathy

Primary GN in India : Post Streptococcal glomerulonephritis

Secondary GN : Diabetes Mellitus.

Most common type of Glomerular Disorder in.

⇒ Leprosy :– MPGN

⇒ Syphilis : Membranous Nephropathy.

⇒ Malaria : Mesangioproliferative.

⇒ Hepatitis C : Cryoglobulinemic glomerulonephritis > membranous glomerulopathy > type 1 MPGN.

⇒ Hepatitis B : Membranous Nephropathy (Component of Hep B virus Responsible – HbsAg).

⇒ SLE – Diffuse glomerulonephritis (class IV) Lupus Nephritis

⇒ Colon cancer / lung cancer – Membranous glomerulopathy.

Rapidly Progressive GlomeruloNephritis

Onset — Has progressed from something.

Type I

Anti GBM antibody

- Renal Limited
- Good Pasture's Syndrome

Type II

Immune Complex mediated

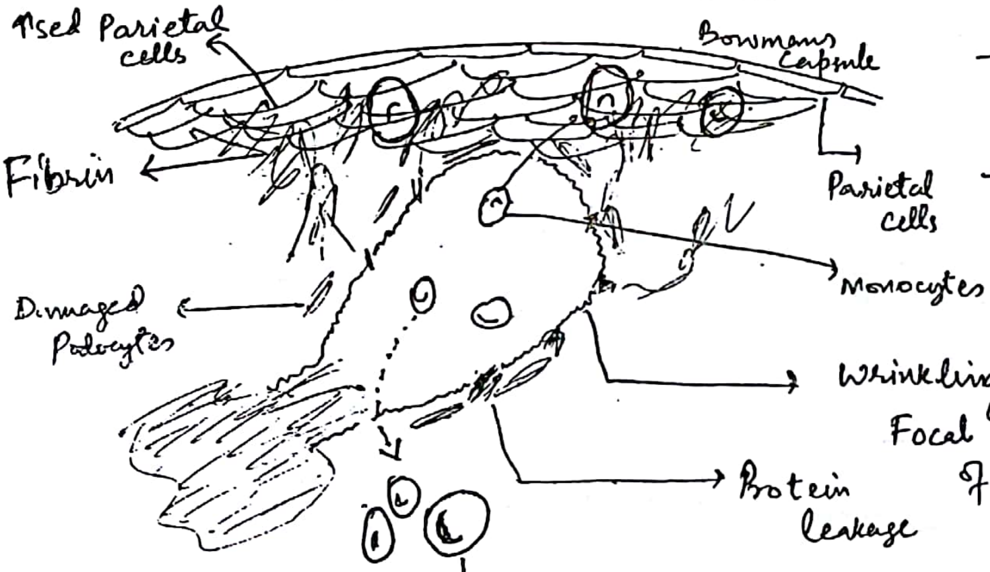
- IgA Nephropathy
- PSGN
- SLE

Type III

ANCA mediated.

(Pauci Immune)

- Wegener's granulomatosis
- CHURG STRAUSS Syndrome
- Microscopic Polyangitis



⇓

⇒ CRECENT ⇒ ↑ Parietal cells, Fibrin, Macrophages

Subepithelial Deposits on EM
+
GRANULAR IF

↓ Nephrotic → Membranous Nephropathy

↓ Nephritic → PSGN.

Hypercellular Glomerulus
+
GRANULAR IF

↓ Subepithelial → Nephritic → PSGN

↓ Subendothelial. → Nephritic + Nephrotic → MPGN

Renal cell Carcinoma

Clear cell | Papillary | Chromophobe

ORIGIN ← Proximal tubule → (Clear cell, Papillary)

Chromophobe: Intercalated cells of C.D.

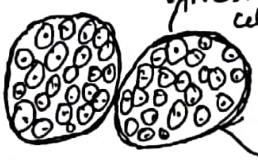
Genetics

Clear cell: 3p deletions → VHL gene

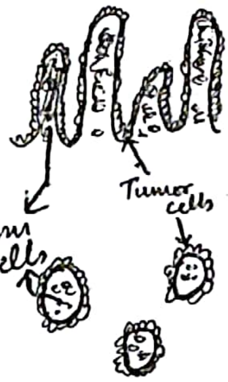
Papillary: Trisomy 7, 17; Loss of Y → ↑ Met-proto-oncogene

Chromophobe: Extreme Hypoploidy.

Histology



Pleomorphism with Clear cytoplasm (because of glycogen/lipid)



- CK7 +ve
- Hale's colloidal iron +ve

Psammoma Bodies (Dystrophic Calcification) [ARISE FROM THE INFARCT and CALCIFICATION OF PAPILLAE TIPS.]

- Papillary Ca thyroid
- Serous cystadenocarcinoma ovary
- Meningioma
- Mesiothelioma

P - Papillary Ca thyroid
P - Prolactinoma
S - Serous CAO
Sa - Somatostatinoma
M - Meningioma
Mo - Mesothelioma

Intercalated cells of collecting duct

↓ gives Rise to

Benign Tumor

$\boxed{\text{Oncocytoma}}$

Angiomyolipoma

Picoma

→ Kidney

→ Manogany brown tumor

→ $\boxed{\text{Central \& Stellate Scar (30\%)}}$

Pancreas

‖

· Central scar.

Histology

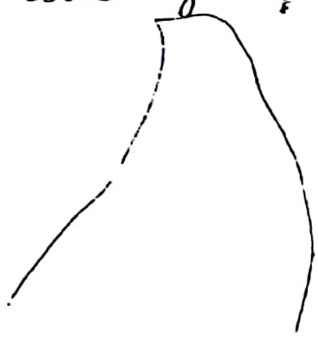
Same as chromophobe except

- No perinuclear halo
- No nuclear pleomorphism

(cells are eosinophilic & granular because of increased number of mitochondria)

Renal cell Carcinoma

- Most common subtype: Clear cell carcinoma
- Most common subtype in sickle cell anemia: Medullary Ca
- Most favourable prognosis: Chromophobe.
- Least favourable prognosis: Sarcomatoid > Medullary > Collecting duct > Clear cell



BREAST PATHOLOGY

Eg - Epithelial hyperplasia
- Atypical hyperplasia
- Insitu carcinoma
- Invasive Carcinoma
- Adenosis
- Small duct papilla

Terminal ductal lobular unit

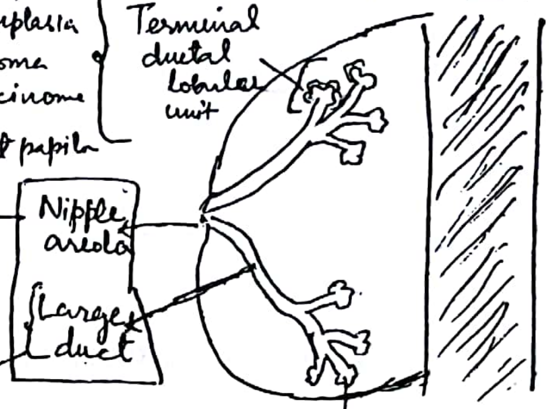
Pectoralis

Paget's disease of Nipple.

Nipple areola

Large duct

Duct ectasia
Duct papilome



Intra lobular strome.

Fibroadenoma
Phyllode's Tumor

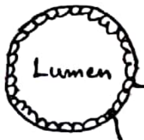
Duct ectasia
(Dilation)

greenish nipple discharge

Superficial erosion

Bloody nipple discharge

Duct papilloma
(Benign papillary tumor of lining Epithelium)

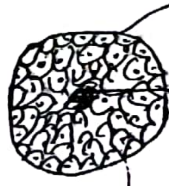


Myoepithelial cells (Basal cells)

Epithelial cells (Luminal cells)

Epithelial hyperplasia
(No pleomorphism of Nuclei)

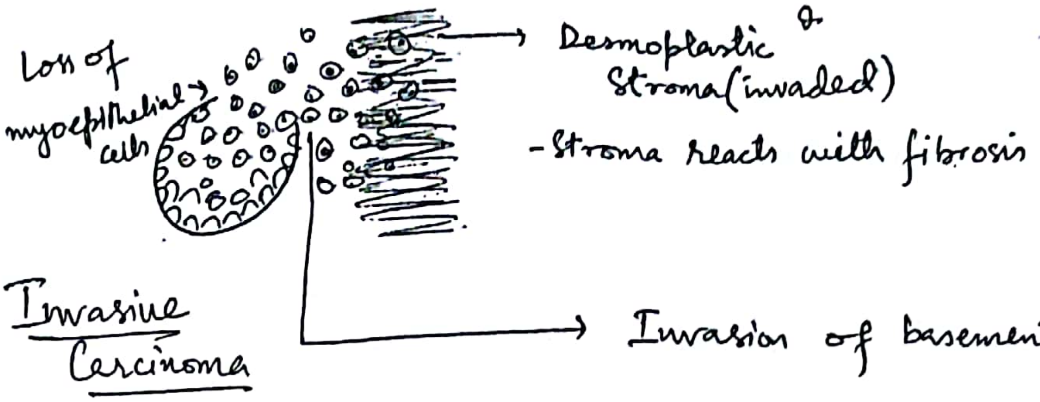
Atypical ductal Hyperplasia



Myoepithelial cell

Central necrosis (comedo DCIS)

Ductal carcinoma in situ (DCIS)



Benign epithelial Lesions

Non Proliferative
(No relative Risk)
- Duct ectasia
- Fibrocystic change
- Apocrine Metaplasia

Proliferative

- Without Atypia (Slight Risk)
  - Duct Papilloma
  - Epithelial Hyperplasia
  - Sclerosing Adenosis
- With Atypia (Moderate Risk)
  - Atypical ductal hyperplasia
  - Atypical lobular hyperplasia

High Risk
- DCIS
- LCIS

Histological Subtypes of Invasive Carcinoma

1. **Invasive Ductal Carcinoma, Not otherwise specified (IDC, NOS)**
   - Most common histological subtype
   - Firm, grey white, Irregular

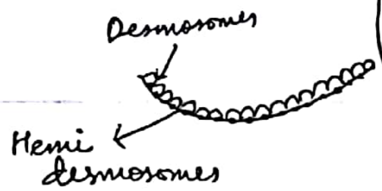
- Grading based on NOTTINGHAM CRITERIA
  - Mitotic activity
  - Tubular differentiation
  - Nuclear pleomorphism.

② Invasive Lobular Carcinoma. (ILC)

CDH 1 gene mutation (Loss of function for making protein E-Cadherin)

↓

$\boxed{\text{E Cadherin loss}}$



Cells become dyscohesive (lose their cohesion)

Become $\boxed{\text{wild \& free!}}$

Single File/Indian File Pattern

⇒ Fail to elicit a desmoplastic response [difficult to detect as a well defined mass on imaging]

⇒ Involve Contralateral Side commonly.

⇓

Metastasize to
- Leptomeningis (arach. + Pia)
- Peritoneum
- Ovaries

③ Medullary Carcinoma

Sheets of tumor cells interspersed with lymphocytes.

↓ Poorly differentiated tumor

↓ Improves treatment response

↓ Thus better prognosis as compared to other poorly differentiated tumors.

Usually associated with BRCA1 gene mutation.

- Triple Negative (ER−, PR−, Her2 Neu−)

④ Tubular Carcinoma

Best prognosis

⑧ Inflammatory Carcinoma

Dermal lymphatics are involved by tumor cells

↓

Peau De orange appearance.

Tumor genetics

Sporadic (more common)[a]

Most common : $\boxed{p53}$

Familial

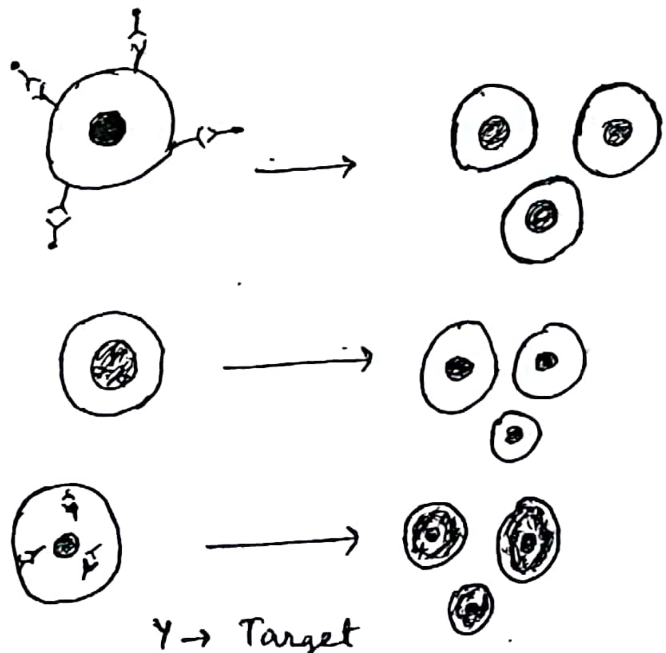
- $\boxed{BRCA1}$ (most common)[a]
  - → Ovarian Cancer
  - Triple Negative (ER–, PR–, Her2Neu–)
- $\boxed{BRCA2}$
  - Male breast Cancer
  - Pancreatic Cancer
  - Prostate Cancer
  - ER+ (Her 2 Neu –ve)
  - ~~Her 2 Neu~~
  - ~~Her 2 Neu~~
- $\boxed{p53}$
  - → Li Fraumeni Syndrome
  - Her 2 Neu +ve
    - ER+
    - ER–

IHC

Chromogen : Di Amino Benzidine (DAB)
BROWN

diamino Benzidine

IHC
⇓
Brown on blue
Test ↗
―――――
Contrast / control



Membranous positivity [Her 2 Neu]

Nuclear Positivity [ER, PR]

Cytoplasmic Positivity [cytokeratin]

Y → Target
Y → Antibody against
• → Chromogen

IHC in Breast Cancer

① **ER/PR**: Allred Score = [0-8]

Intensity + % of positive cells.

② **Her-2-Neu**

% of cells positive & intensity.

- 0 +1 → Negative
- +2 → **Equivocal**
  - **Perform FISH** (To look for Her 2 Neu Amplification)
    - Negative
    - Equivocal → Repeat testing on other sample
    - Positive Amplified → TRANSTUZUMAB Therapy
- +3 → Positive → TRANSTUZUMAB Therapy

Breast Carcinoma

Most common histological Subtype – $\boxed{\text{IDC, NOS}}$

Most common molecular subtype – Luminal A

Histological subtype with Best prognosis – Tubular > Mucinou

Molecular subtype with Best prognosis – Luminal A

Most common genetic mutation – $\boxed{p53}$.

Most common familial genetic mutation – BRCA 1.

Highest susceptibility seen with which mutation – BRCA 1

Association of male breast cancer, prostate cancer, pancreatic Ca, melanoma and gastric Cancer – BRCA 2

Association of ovarian cancer (serous) – BRCA1 > BRCA2

Most common molecular group associated with BRCA1 – $\boxed{\text{BAX}}$

Most common molecular group associated with BRCA2 – Luminal B

Most important prognostic marker – Lymph node status.

Most important marker (prognostic) for metastatic Cancer – ER/PR status

Least desmoplastic tumor – Lobular Carcinoma.